ABSTRACT

PHILLIP CRUNK. Physical and Chemical Evaluation of Continuous Thermophilic Anaerobic Digestion of Wastewater Sludge. (Under the direction of Dr. Michael D. Aitken)

The goal of this study was to evaluate the performance of a completely-mixed flow reactor (CMFR) used to treat municipal wastewater sludge under varied thermophilic operating temperatures (55, 53, and 51 °C) and at low a hydraulic retention time (HRT). Parameters investigated included pH, alkalinity, total and volatile solids destruction, residual volatile fatty acid (VFA) concentrations, gas production, gas composition, and ammonium nitrogen (NH_4-N) concentrations. This study was part of a larger investigation as an attempt to obtain approval from the U.S. Environmental Protection Agency (EPA) for a unique thermophilic anaerobic reactor configuration developed by the South Columbus (Georgia) Water Resources Facility (SCWRF) and Brown and Caldwell, Inc.. The bench-scale study coupled a CMFR with a completely-mixed batch reactor (CMBR) to meet pathogen criteria inactivation established by the EPA for Class A municipal biosolids.

TABLE OF CONTENTS

LIST OF TABLES

LIST OF FIGURES

1. INTRODUCTION

Wastewater treatment facilities are increasingly turning to the application of treated wastewater solids onto land as a disposal solution due to recent flexibility in land application regulations (National Research Council Committee on Toxicants and Pathogens in Biosolids Applied to Land, 2002). In fact, 60% of the 5.6 million dry tons of sewage sludge produced in the United States is used for land application (National Research Council Committee on Toxicants and Pathogens in Biosolids Applied to Land, 2002).

Disposal of biosolids produced in the treatment of municipal wastewater sludge is regulated by the U.S. Environmental Protection Agency (EPA) under 40 CFR Part 503. With the 1993 promulgation of Part 503, quality-based criteria in addition to the pre-existing technology-based criteria offer more versatility in achieving safe levels of sludge treatment. Depending on the extent of treatment, biosolids intended for land application are designated as either Class A or Class B. Class A biosolids are essentially free of pathogenic microorganisms and there are six alternative sets of criteria by which biosolids are considered to have met the Class A criteria. Sludge treatment processes designated as "processes to further reduce pathogens" (PFRP) are considered to produce Class A biosolids if specified operating conditions are met, but thermophilic anaerobic digestion (TAD) is not listed as a PFRP in the Part 503 regulations. Therefore an alternative criterion must be met if biosolids produced from TAD are to be designated Class A.

Alternative 1 of the Class A biosolids criteria specifies durations of treatment based on temperature and feed solids concentration. Alternative 1 requirements apply to batch or plug-flow reactors, but exclude mixed, continuous-flow digesters because of the potential for short-circuiting of the influent sludge (U.S. Environmental Protection Agency, 1999). Since mesophilic, anaerobic, mixed continuous or semi-continuous-flow reactors are most commonly employed by wastewater treatment plants for sludge treatment, simply converting conventional mesophilic digesters to thermophilic operating temperatures may not meet the Class A criteria. Under Alternative 6, biosolids can meet the Class A criteria by demonstrating that the sludge treatment process is equivalent to a PFRP. Equivalency requests are evaluated by EPA's Pathogen Equivalency Committee (PEC). Class A equivalency can be sought and granted for either a specific treatment plant (site-specific equivalency) or for a generic process (national

equivalency).

Columbus Water Works (Columbus, GA) and Brown and Caldwell, Inc. have developed an approach to achieving the Class A criteria with TAD called the Columbus Biosolids Flow-Through Thermophilic Treatment ($CBFT^3$) process. The $CBFT^3$ process combines a mixed, continuous-flow reactor followed by a nominally plug-flow reactor or a batch detention step with a retention time that is much shorter than that required from the relevant Part 503 time-temperature equation. The second reactor would compensate for any short-circuiting that might occur in the mixed, continuous-flow reactor. The pathogen indicator organism inactivation data resulting from the operation of this bench-scale reactor configuration can be reference in an unpublished paper submitted to Brown and Caldwell, Inc. in Atlanta, GA (Aitken et al., 2003). National PFRP equivalency is being sought for the $CBFT^3$ process through a demonstration project. The demonstration project includes a laboratory investigation, full-scale prototype evaluation of the plug-flow reactor, and a full-scale demonstration at the South Columbus (GA) Water Resources Facility (SCWRF).

The objective of this study was to evaluate the effect of short residence times (4-6 days), a range of operating temperatures (55, 53, 51 °C), and diverse source sludges on the performance of the continuous laboratory digester. The physical and chemical parameters that were evaluated were pH, alkalinity, volatile fatty acid concentrations (VFAs), ammonium nitrogen concentrations, total and volatile solids, and gas composition and production. These physical and chemical parameters indicate levels of stability and stabilization efficiency at which the CMFR was performing.

2. LITERATURE REVIEW

2.1 Wastewater treatment residuals

The sources of municipal wastewater and method of treatment determine the characteristics of wastewater solids that must be treated and/or disposed. In conventional secondary treatment plants, the solid material that settles out of wastewater within the primary and secondary sedimentation basins are termed primary and waste activated sludge or biosolids, and both make up the majority of solids that are either stabilized, incinerated or landfilled by the reclamation facility (Metcalf and Eddy, Inc., 2003). Common sludge characteristics and their dependence on the treatment process can be seen below in Table 2.1.

Untreated sludges are hazardous both because of their high pathogenic concentrations and because they attract vectors of disease such as insects, birds, and rodents. Pathogen types found in municipal sludges include bacteria, viruses, protozoa, and parasites (U.S. Environmental Protection Agency, 1994). Pathogen concentrations in municipal sludge are a function of demographics, but enterovirus concentrations of primary sludges commonly range from 7,000 to 10,000 and waste activated biosolids typically contains less than 500 plaque-forming units (pfu) per kg of solids (Schwartzbrod, 1995). The most important global impact of untreated wastewater and wastewater solids are diseases caused by parasitic intestinal worms or helminths (Cabirol et al., 2002). In Mexico *Ascaris* helminths infect 43 to 94% of rural populations (Cabirol et al., 2002).

Along with pathogen inactivation the other goal of sludge treatment is reducing vector attraction, which is quantified by the EPA in units of reduced volatile solids or chemical oxygen demand (U.S. Environmental Protection Agency, 1994). Reducing pathogens and vector attraction are two key objectives in treating wastewater sludges, the extent to which is determined by the intended use or nonuse of the product.

Table 2.1. Municipal Sludge Characteristics (Metcalf and Eddy, Inc., 2003)

Product	Specific gravity of solids	Specific gravity of sludge	% Dry Solids Concentration	
			Range	Typical
Conventional primary sludge	1.4	1.02	5-9	6
Primary sludge with high lime addition for phosphorus removal	2.2	1.05	4-16	10
Conventional waste activated biosolids after primary settling	1.25	1.005	0.15-1.5	0.8
Waste activated biosolids with high-purity oxygen and primary settling	-	-	1.3-3	2
Conventional primary sludge and waste activated biosolids	-	-	3-8	4

2.2 Land Application

2.2a Soil amendments

Treated wastewater sludges, termed biosolids, are often applied to land as soil amendments. Biosolids have been used to enhance agricultural soils, forest, recreational areas, and reclaimed Superfund sites and strip mines (Metcalf and Eddy, Inc., 2003). Over half of the biosolids produced in the U.S. are used for land application (National Research Council Committee, 2002). Biosolids provide a multifaceted soil enhancement (Page et al., 1987).

Treated wastewater sludges contribute macronutrients such as nitrogen and phosphorus, as well as micronutrients such as iron and manganese (Metcalf and Eddy, Inc., 2003). In fact, biosolids protect against nutrient rich runoff or leachate, because the organic nutrients in biosolids are less soluble and are released more slowly than conventional fertilizers (Page et al., 1987). The organic matter found in biosolids also improves the cation-exchange capacity of soil, allowing retention of potassium, calcium, and magnesium (Metcalf and Eddy, Inc., 2003).

Along with chemical improvements, additional organic matter in biosolids also increases biodiversity and activity in the soil, loosening clay and improving sandy soils (Page et al., 1987). Land application reduces soil erosion and enhances water retention, tilth, infiltration, and

aeration encouraging root growth and making crops more drought resistant (Metcalf and Eddy, Inc., 2003). Biosolids improve soils physically, chemically, and biologically.

2.2b Pathogen viability in soils

Despite the benefits of land-applied sludges, without proper treatment the survival of pathogens can pose a persistent public health threat. Helminths are a primary concern in land-applied sludges, because of their persistence in soils (Helmer and Hepanhol, 1997). Provided the needed nutrients and moisture, helminth ova can remain viable for a span from months to years in soils (Helmer and Hepanhol, 1997). Enteric viruses also have a persistent life span (Schwartzbrod, 1995). Table 2.2 summarizes important wastewater pathogens and their life spans in soils and plants.

Table 2.2. Survival Times for Pathogens in Soils and on Plant Surfaces (U.S. Environmental Protection Agency, 1999)

	Soil		Plants	
Pathogen	Absolute Maximum	Common Maximum	Absolute Maximum	Common Maximum
Bacteria	1 Year	2 months	6 months	1 month
Viruses	1 Year	3 months	2 months	1 month
Protozoan cysts	10 days	2 days	5 days	2 days
Helminth ova	7 years	2 years	5 months	1 month

Environmental factors that affect the viability of viruses include ultraviolet exposure, presence of aerobic microbes, pH, temperature, humidity, and adsorption onto soil particles (Schwartzbrod, 1995). Low temperatures and high humidity are conducive to extended viral life spans (Schwartzbrod, 1995).

2.2c Pathogen migration in soils

Along with varied life spans, different pathogens also migrate through the subsurface differently (Schwartzbrod, 1995). The potential for migration converts both agricultural

products and groundwater into possible pathways for exposure to pathogens (Schwartzbrod, 1995). Most pathogens remain near the surface of the soil, in which case tubers such as potatoes, carrots, and radishes may accumulate hazardous pathogenic concentrations (Schwartzbrod, 1995). However, viruses can adsorb and desorb from soil particles, as well as infiltrate through the soil matrix (Schwartzbrod, 1995). Adsorption depends on soil and virus type, and some studies disagree on how adsorption affects viability (Schwartzbrod, 1995). Heavy rain and irrigation, seepage kinetics, and water table depths are key variables in assessing the risk level of groundwater contamination (Schwartzbrod, 1995). The movement and persistence of pathogens make land application of untreated or inadequately treated biosolids potentially dangerous if the land is used in a manner that can lead to human exposure.

2.2d Regulatory history

The EPA began promoting the recycling of biosolids in the early 1970's (National Research Council Committee on Toxicants and Pathogens in Biosolids Applied to Land, 2002). The 1977 amendments to the Clean Water Act (CWA) and the 1976 Resource Conservation and Recovery Act (RCRA) required federal regulators to draft future legislation addressing responsible treatment and disposal of biosolids (U.S. Environmental Protection Agency, 1999). After 1979, Title 40 Part 257 established technology-based requirements to regulate the use and disposal of wastewater sludge (U.S. Environmental Protection Agency, 1995). In 1988 the Ocean Disposal Ban Act prohibited the dumping of biosolids offshore and increased the need for alternatives to biosolids disposal (National Research Council Committee on Toxicants and Pathogens in Biosolids Applied to Land, 2002). Finally on February 19, 1993, 40 CFR Part 503, Biosolids Rule, was promulgated (U.S. Environmental Protection Agency, 1999).

2.2e Current regulatory status

The current 503 regulation is a quality-based regulation that defines criteria for Class A and Class B biosolids, as well as the approved technologies already established in Part 257.

Part 503 offers alternatives to achieving pathogen criteria regarding Class A or B biosolids production versus Part 257, which only provided limited lists of approved technologies (PFRP, PSRP) for meeting Class A or B criteria (U.S. Environmental Protection Agency, 1994). The approved alternatives for producing Class A biosolids are included below in Table 2.3.

Table 2.3. Six Alternatives for Meeting Class A Pathogen Requirements (U.S. Environmental Protection Agency, 1994)

Alternative 1: ***Thermally Treated Biosolids***
Biosolids must be subjected to one of four time-temperature regimes.
Alternative 2: ***Biosolids Treated in a High pH-High Temperature Process***
Biosolids must meet specific pH, temperature, and air-drying requirements
Alternative 3: ***Biosolids Treated in Other Processes***
Demonstrate that the process can reduce enteric viruses and viable helminth ova. Maintain operating conditions used in the demonstration after pathogen reduction demonstration is completed.
Alternative 4: ***Biosolids Treated in Unknown Processes***
Biosolids must be tested for pathogens-*Salmonella* sp. or fecal coliform bacteria, enteric viruses, and viable helminth ova-at the time the biosolids are used or disposed, or, in certain situations, prepared for use or disposal.
Alternative 5: ***Biosolids Treated in a PFRP***
Biosolids must be treated in one of the Processes to Further Reduce Pathogens (PFRP).
Alternative 6: ***Biosolids Treated in a Process Equivalent to a PFRP***
Biosolids must be treated in a process equivalent to one of the PFRPs, as determined by the permitting authority.

Alternative 1 refers to four equations that calculate required batch treatment contact times as a function of temperature, solids concentration, and method of treatment (U.S. Environmental Protection Agency, 1999). Alternative 6 states that the Pathogen Equivalency Committee (PEC) can approve technologies for achieving inactivation and stabilization equivalent to a PFRP (U.S. Environmental Protection Agency, 1999). For comparison, the Class B alternatives are also included below (Table 2.5). Alternative 1 for producing Class B biosolids identifies the indicator organism and the allowable concentration. Alternative 2 references the PSRP list of technologies, under which TAD is currently classified (U.S. Environmental Protection Agency, 1999).

The distinction between Class A and B biosolids is the allowable concentrations of pathogenic organisms. All six of the established alternatives for producing Class A biosolids

hinge on their capacity to inactivate specified indicator organisms to non-detectable concentrations (U.S. Environmental Protection Agency, 1994). Table 2.4 details the indicator organisms and the inactivation requirements that all six alternatives Class A biosolids must be able to achieve.

Table 2.4. Pathogen Requirements for All Class A Alternatives (U.S. Environmental Protection Agency, 1999)

Pathogen	Criteria
Salmonella sp.	less than 3 MPN per 4 grams total solids biosolids (dry weight basis)
Enteric viruses	less than 1 PFU per 4 grams total solids biosolids (dry weight basis)
Viable helminth ova	less than 3 MPN per 4 grams total solids biosolids (dry weight basis)

In contrast to Class A pathogen criteria, the Class B inactivation requirements not as stringent, which would explain the multiple restrictions applied areas in which Class B biosolids are used (U.S. Environmental Protection Agency, 1994). The allowable indicator organism concentrations are included in Alternative 1 of the Class B pathogen requirements found in Table 2.5.

The vector attraction requirements are the same for both Class A and B biosolids, and serve to ensure there are sufficient reductions in organic content, odor, and pathogen regrowth potential (U.S. Environmental Protection Agency, 1994). Odor and pathogen regrowth are important factors that can lead to the spread of diseases through vectors such as insects, rodents, and birds (U.S. Environmental Protection Agency, 1994). A list of the 12 options for demonstrating sufficient vector attraction reduction is included in Table 2.6.

Table 2.5. Three Alternatives for Meeting Class B Pathogen Requirements (U.S. Environmental Protection Agency, 1994)

Alternative 1: ***The Monitoring of Indicator Organisms***
Test for fecal coliform density as an indicator for all pathogens. The geometric mean of seven samples shall be less than 2 million MPN per gram per total solids or less than 2 million CFU per gram of total solids at the time of use or disposal.
Alternative 2: ***Biosolids Treated in a PSRP***
Biosolids must be treated in one of the Processes to Significantly Reduce Pathogens (PSRP).
Alternative 3: ***Biosolids Treated in a Process Equivalent to a PSRP***
Biosolids must be treated in a process equivalent to one of the PSRP, as determined by the permitting authority.

Table 2.6. Summary of Options for Meeting Vector Attraction Reduction (U.S. Environmental Protection Agency, 1994)

Option 1: Meet 38 percent reduction in volatile solids content.
Option 2: Demonstrate vector attraction reduction with additional anaerobic digestion in a bench-scale unit.
Option 3: Demonstrate vector attraction reduction with additional aerobic digestion in a bench-scale unit.
Option 4: Meet a specific oxygen uptake rate for aerobically digested biosolids.
Option 5: Use aerobic processes at greater than 40 °C for 14 days or longer.
Option 6: Alkali addition under specified conditions.
Option 7: Dry biosolids with no unstabilized solids to at least 75 percent solids.
Option 8: Dry biosolids with unstabilized solids to at least 90 percent solids.
Option 9: Inject biosolids beneath the soil surface.
Option 10: Incorporate biosolids into the soil within 6 hours of application to or placement on the land.
Option 11: Cover biosolids placed on a surface disposal site with soil or other material at the end of each operating day. (Note: Only for surface disposal.)
Option 12: Alkaline treatement of domestic septage to pH 12 or above for 30 minutes without adding more alkaline material.

2.3 Stabilization Processes

In order to decrease the potential health risks and problems in disposing of wastewater sludges, varied methods of stabilization have been developed to attenuate the organic matter

resulting from wastewater treatment. Wastewater sludges are stabilized to reduce pathogens, offensive odors, and the potential for putrefaction (Metcalf and Eddy, Inc., 2003). A few examples of stabilization methods are lime stabilization, composting, heat- or air-drying, irradiation, pasteurization, and aerobic or anaerobic digestion (U.S. Environmental Protection Agency, 1994). All of these methods of stabilization inactivate pathogens and reduce putrefaction potential to varying degrees of efficacy. Each method also has disadvantages in terms of increasing the ultimate volume for disposal, the time required, the space required, or the energy required. The disadvantages of these stabilization methods relative to their advantages are the reasons why they are not the most commonly used in municipal sludge treatment.

2.3a Anaerobic digestion

One of the oldest and most widely implemented stabilization processes is anaerobic digestion (Metcalf and Eddy, Inc., 2003). In fact worldwide, stabilization of wastewater sludge is the most common application of anaerobic digestion, because it is a highly efficient method of stabilization for most wastewater treatment plants (Ahring et al., 2002; Oles et al., 1997). Along with pathogen inactivation and volatile solids reduction, anaerobic digestion reduces product volume, improves the dewaterability of the product, and produces methane gas (Oles et al., 1997). Anaerobic digestion does not require energy for aeration and offers the potential for recovering energy from the methane production, making it an energy-efficient option for treatment facilities (Metcalf and Eddy, Inc., 2003). The term anaerobic digestion describes a series of microbial reactions used to decompose organic and some inorganic matter such as sulfate (Metcalf and Eddy, Inc., 2003). Different environmental conditions can support the various microbial consortia capable of completing the desired metabolic reactions. Hence, there are several approaches to anaerobic digestion used in practice.

The various methods of anaerobic digestion are defined by differences in environmental conditions such as temperature, feeding rate, and feeding method. The most encompassing variable is temperature, because of the different microbial populations that are cultivated under different operating temperatures. Typically, anaerobic digesters operate within two temperature ranges, mesophilic (25 to 42 ^{0}C) or thermophilic (45 to 65 ^{0}C) (Schafer et al., 2003). Conventionally, mesophilic anaerobic digestion is the most commonly employed stabilization method (Gabb et al., 2001). Temperature-phased anaerobic digestion, which is practiced by

about a dozen U.S. facilities, combines both mesophilic and thermophilic processes in series, and offers the product qualities inherent at both temperatures (Schafer et al., 2003). Other distinctions among different methods of anaerobic digestion involve varied feeding rates that are increased or decreased based on the desired metabolic activity (Schafer et al., 2003). Acid-gas phased digestion makes use of this technique, providing higher feeding rates for the acetogenic organisms in the first step of the process (acid-phase reactor) and subsequently providing lower feeding rates for the methanogenic organisms in a second reactor (gas-phase or methanogenic reactor) (Schafer et al., 2003).

2.3b Thermophilic anaerobic digestion (TAD)

Despite the conventional dominance of mesophilic anaerobic digestion, thermophilic temperatures offer a multitude of advantages, and recent regulation has encouraged research into and implementation of TAD (Gabb et al., 2001). Some parts of the world already rely primarily on TAD. Denmark, for example, used mesophilic anaerobic digestion predominantly since the 1950's, but by 1995 70% of the anaerobic digesters operated in the thermophilic temperature range (Nielsen and Petersen, 2000).

As a rule of thumb, biochemical rates approximately double with an increase of 10 °C until asymptotic rates are neared (Metcalf and Eddy, Inc., 2003). Therefore, a fundamental benefit of TAD is an increase in growth rates and metabolic rates that enable further stabilization, enhanced gas production, improved dewatering, lower retention times, and more pathogenic inactivation than comparable mesophilic operations (Parkin and Owen, 1986; Gabb et al., 2001; Buhr and Andrews, 1977; Zabranska et al., 2000).

The inherent advantages to thermophilic metabolism can translate into more efficient stabilization unit processes. Technological advancements have helped outdate the association of thermophilic digestion with higher energy demands. Increased methane production coupled with improved technology in dewatering, energy recovery, and heat exchangers have enabled thermophilic operations to become self-sustaining if not energetically profitable (Garber, 1982; Rimkus et al., 1982; Zabranska et al., 2002; Metcalf and Eddy, Inc., 2003).

The potential for decreased retention times for equivalent stabilization translates into the need for smaller reactor volumes for thermophilic digestion relative to mesophilic digestion, allowing for lower construction cost or additional existing space for expansion. In some cases

retention times have been reduced in half and digester capacities doubled (Nielsen and Petersen, 2000; Zabranska et al., 2002).

Enhanced dewaterability of the biosolids produced is another well-documented benefit of TAD. In a comparison of the filterability of mesophilic and thermophilic biosolids resulted in the thermophilic product had a four-fold higher rate of filtration (29.3 $kg/m^2/h$ vs 7.3 $kg/m^2/h$, respectively), also decreasing the energy demand by 40% and requiring only half the amount of polymer (Garber, 1982). In the same study the thermophilic cake density was 10% greater than the mesophilic cake density (Garber, 1982). Initial volumes of raw sludge can be reduced by 30-40% due to TAD's capacity for high solids reduction and the enhanced dewaterability, which is important when considering the costs of landfilling (Nielsen and Petersen, 2000).

With regard to the most recent biosolids regulations, the most valuable characteristics of thermophilic temperatures are the accelerated rates of pathogen inactivation (Grady et al., 1999). Although TAD is not classified as a PFRP, the process has been proven capable of faster pathogen inactivation rates than would be indicated by the time-temperature requirements of the Part 503 regulations (Grady et al., 1999; Schwartzbrod, 1995; Gabb et al., 2001; Cabirol et al., 2002).

2.4 Microbiological Processes in Anaerobic Digestion

Anaerobic digestion is a multi-phase biological process with all phases occurring simultaneously and, in most cases, interdependently. The balance among these steps dictates digester performance and stability. The first step is hydrolysis and liquefaction where acidogenic bacteria convert complex organics to more soluble products (Parkin and Owen, 1986; Grady et al., 1999). In the second step of the process, the hydrolyzed organics are converted by acidogens to VFAs, such as acetic, propionic, and butyric acids, as well as hydrogen gas (Grady et al., 1999). In subsequent steps the resulting short-chain VFAs (propionic, butyric, etc.) and long-chain fatty acids are anaerobically oxidized by acetogens to form acetic acid and hydrogen (H_2) (Parkin and Owen, 1986; Grady et al., 1999). Finally the last two steps involve the ultimate formation of methane (CH_4) by either acetoclastic methanogens or H_2-oxidizing (CO_2-reducing) methanogens (Parkin and Owen, 1986; Grady et al., 1999). Anaerobic digestion therefore encompasses the simultaneous activity of five major groups of microorganisms: fermentative bacteria, hydrogen-producing acetogenic bacteria, hydrogen-consuming acteogenic bacteria, H_2-

oxidizing methanogens, and acetoclastic methanogens (Parkin and Owen, 1986). A flow chart of anaerobic digestion and a proportional pathway of chemical oxygen demand (COD) are provided in Figures 2.1 and Figure 2.2.

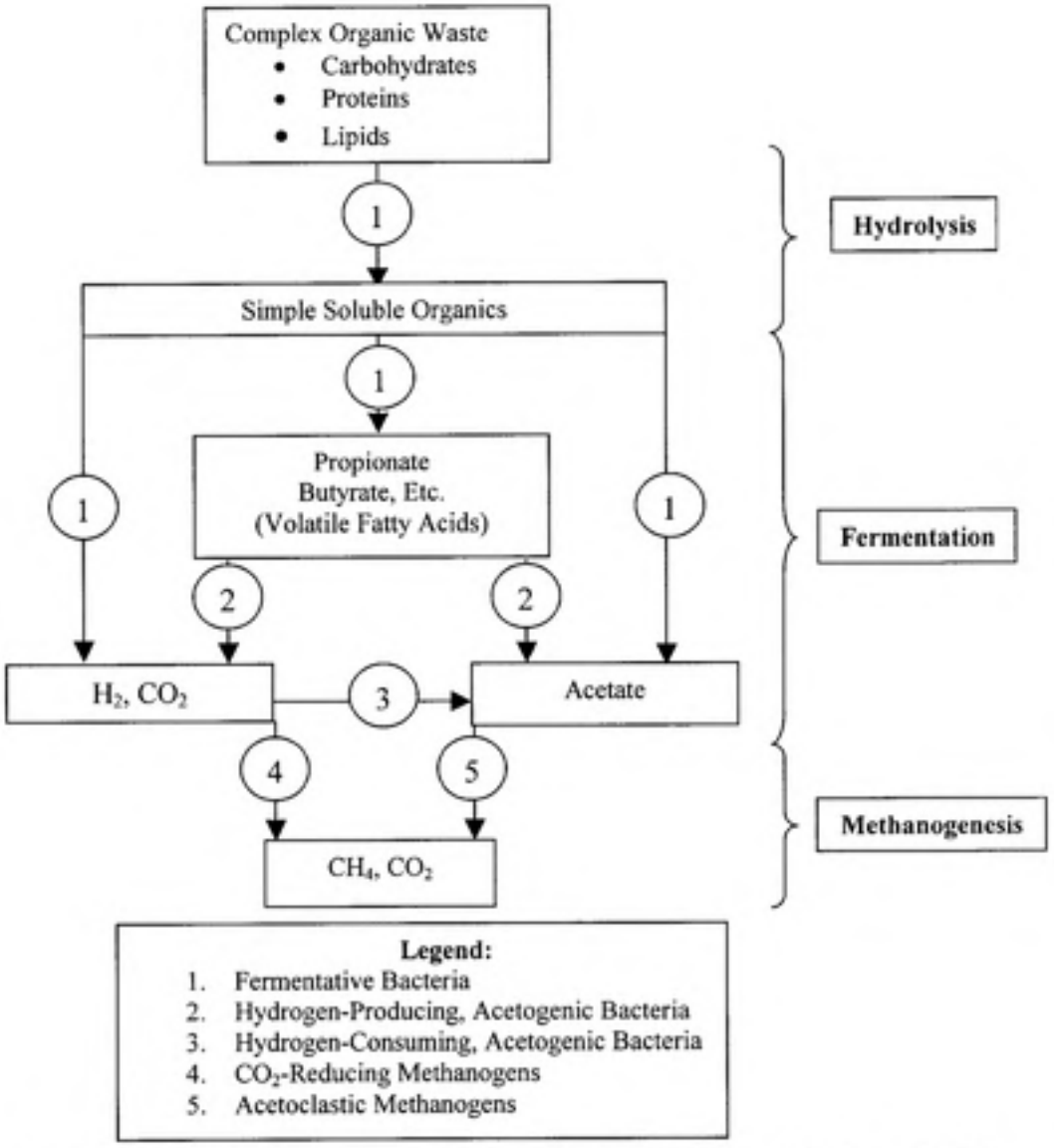


Figure 2.1. Flow chart of anaerobic digestion (Grady et al., 1999; Parkin and Owen, 1986)

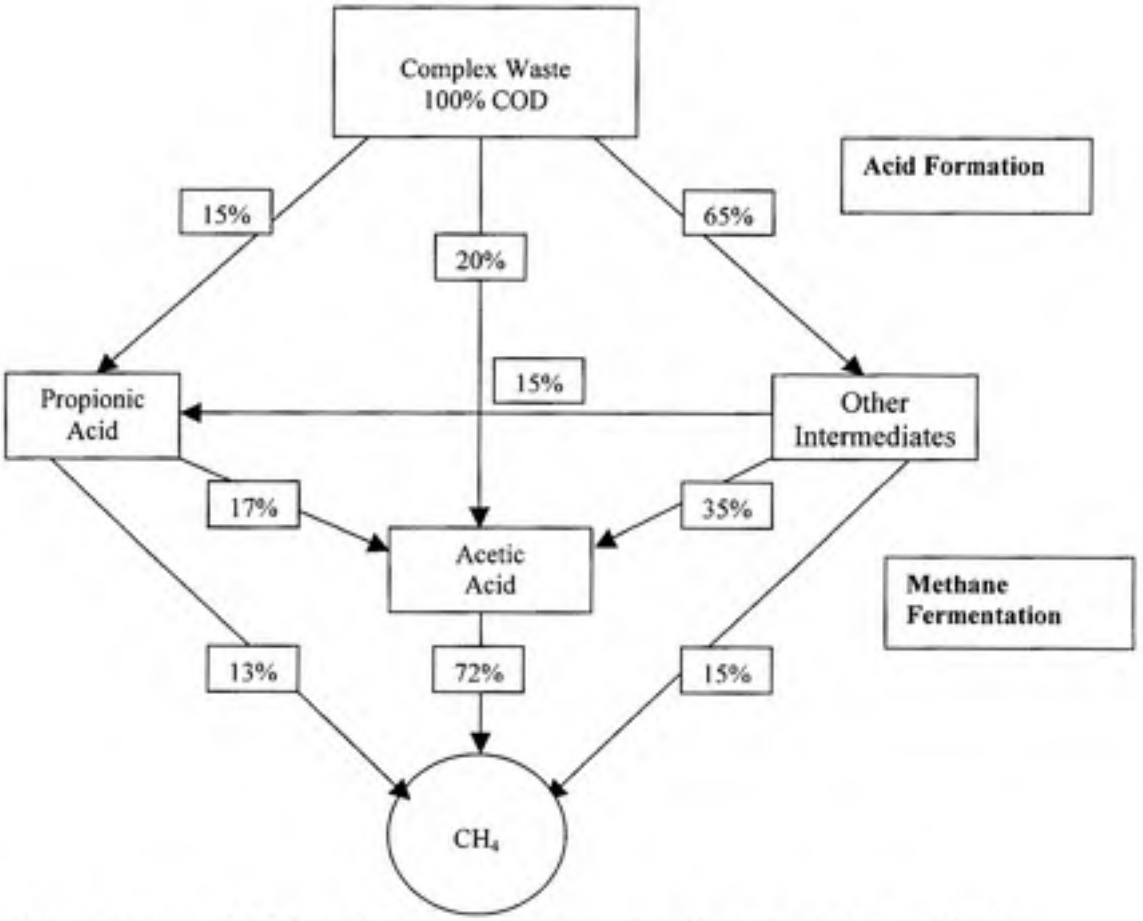


Figure 2.2. Pathway of COD during Anaerobic Digestion (Parkin and Owen, 1986)

2.4a Hydrolysis

Hydrolysis and then fermentation are both performed by the fermentative bacteria that are either facultative or obligate anaerobes (Parkin and Owen, 1986). During hydrolysis and liquefaction, complex and/or insoluble organics such as carbohydrates, proteins, and lipids are converted to simpler, more soluble forms that can pass through bacterial cell walls and membranes to be used as energy sources (Parkin and Owen, 1986). This step is particularly important for proportionally large, insoluble components of municipal feed sludges such as cellulose and lipids (Parkin and Owen, 1986). Hydrolysis is accomplished with extracellular enzymes such as lipases, cellulases, amylases, and proteases (Grady et al., 1999). Extracellular enzymes are excreted by the bacteria, making contact time and complete mixing critical operating parameters to ensure hydrolysis is not the limiting step in stabilization (Parkin and

Owen, 1986). The most likely rate-limiting step in hydrolysis is the conversion of lipids (Grady et al., 1999).

2.4b Fermentation and acetogenesis

Once the complex organics are hydrolyzed into simple carbohydrates, amino acids, and long chain fatty acids, fermentative bacteria (acidogens) use those compounds for energy in producing hydrogen, carbon dioxide, acetate, and short-chain or long-chain volatile fatty acids (VFAs) such as propionic, butyric, and valeric acids (Grady et al., 1999; Metcalf and Eddy, Inc., 2003; Rittmann and McCarty, 2001). Acetogens use the higher-molecular-weight VFAs in the formation of acetic acid, with the conversion of propionic acid a potential rate-limiting step (Griffin et al., 1998; Grady et al., 1999; Ahring et al., 2001; Kim and Speece, 2002). The H_2 production by fermentative bacteria is a relatively small contribution to total H_2 production (Grady et al., 1999). The majority of H_2 is produced by anaerobic oxidation of long- and short-chain fatty acids to acetic acid (Grady et al., 1999). The production of hydrogen is important because it is the substrate for hydrogen-oxidizing methanogens (Grady et al., 1999). As H_2 escapes from solution into the gas phase it serves as an electron sink, making it thermodynamically conducive for acetate formation (Grady et al., 1999). If not for the formation of H_2 then VFAs with molecular weights greater than that of acetate would accumulate and methanogenesis would not be possible (Parkin and Owen, 1986, Grady et al., 1999). In summary, fermentation and acidogenesis involve the conversion of hydrolyzed organics to produce primarily acetate, hydrogen, and carbon dioxide, with other VFAs as the intermediary compounds (Grady et al., 1999).

2.4c Methanogenesis

Methanogenesis refers to the conversion of carbon to its most reduced state, methane (CH_4) (Rittmann and McCarty, 2001). Methanogenesis involves two types of methanogens that convert the hydrogen, carbon dioxide, and acetic acid produced during the previous phases into methane and carbon dioxide gases (Parkin and Owen, 1986). Acetate fermenters produce methane and carbon dioxide from the cleavage of acetate, while hydrogen oxidizers produce methane from hydrogen and carbon dioxide (Rittmann and McCarty, 2001). Acetate cleavage represents 72% of the methane production in the typical sludge digesters (Parkin and Owen,

1986). However, compared to hydrogen oxidizers, acetate fermenters have a very low energy yield (0.45 g VSS/ g H_2 vs 0.04 g VSS/g Ac, respectively) (Rittmann and McCarty, 2001). Due to their slow growth rate, acetate fermenters and propionic acid degraders are probable rate-limiting species in anaerobic digestion (Parkin and Owen, 1986; Rittmann and McCarty, 2001). Carbon dioxide reduction associated with H_2 oxidation accounts for the remaining 28% of the methane production (13% of the COD derived from propionic acid and 15% from other intermediates) (Parkin and Owen, 1986). The volatilization of the gaseous methane and carbon dioxide from solution is the defining step in waste stabilization (Rittmann and McCarty, 2001). To summarize, the third phase of digestion is carried out by acetoclastic and CO_2-reducing methanogens that utilize acetic acid, hydrogen, and carbon dioxide in producing methane (Parkin and Owen, 1986). A net production of carbon dioxide results from the acetoclastic methanogenesis.

2.5 Performance Parameters

Performance parameters indicate the stability and efficiency of the digestion process. These parameters include pH, alkalinity, total and volatile solids, ammonium-nitrogen, gas production and composition, and volatile fatty acids. Common ranges of stability, critical reactions, effects of operating parameters, and interpretation of these performance parameters are addressed in this section.

2.5a pH

As in all biochemical reactions, many important factors determine and are determined by pH, such as chemical equilibria and pending stability (Grady et al., 1999). In general a pH between 6.8 and 7.6 is optimal for TAD (Parkin and Owen, 1986; Metcalf and Eddy, Inc., 2003).

Operating parameters that influence pH are temperature and residence time. Increasing temperatures decrease the solubility of CO_2, which can drive the pH upward (Buhr and Andrews, 1977). Conversely, shorter residence times can also drive the pH downward because slower-growing methanogen populations decrease, which results in increasing VFA concentrations. (Buhr and Andrews, 1977; Garber, 1982).

One inhibitory factor to digestion that is pH-dependent is the ammonia (NH_3) concentration, the deprotonated form of ammonium (NH_4^+). Optimal methanogenic activity is

achieved at a pH between 7.0 and 7.5 (Liu and Sung, 2002). More basic or higher pH environments result in the conversion of ammonium (NH_4^+) to ammonia (NH_3), a process described with the following equilibrium relationships at 25 °C (Liu and Sung, 2002; Dean, 1985).

$$NH_4^+{}_{(aq)} \leftrightarrow NH_{3(aq)} + H^+ \qquad K_a = [NH_{3(aq)}][H^+]/[NH_4^+{}_{(aq)}] = 10^{-9.24} \text{ (mole/L)} \qquad (1)$$

$$NH_{3(g)} \leftrightarrow NH_{3(aq)} \qquad K_H = [NH_{3(aq)}]/[NH_{3(g)}] \text{ (mole/L)} \qquad (2)$$

Acidic or low pH environments can also indicate disproportionate microbial populations and are potentially inhibitive. Generally, a pH below 6.8 can inhibit methanogenesis (Metcalf and Eddy, Inc., 2003). Acidic conditions are indicative of an imbalance in the interspecies transfer of hydrogen between the methanogenic hydrogen consumers, and the acidogenic hydrogen producers (Metcalf and Eddy, Inc., 2003). Studies have shown that when the partial pressure of H_2 exceeds 10^{-4} atm, reactions responsible for converting the higher-molecular weight VFAs to acetate are thermodynamically unfavorable, leading to the accumulation of these VFAs. Methanogenesis is inhibited as a result, since these VFAs are not substrates for methanogens (Grady et al., 1999; Parkin and Owen, 1986). Evidence suggests that acidogens replicate at a maximum specific growth rate one order of magnitude higher than methanogens (Ghosh, 1987). Therefore, single stage anaerobic digesters operating below critical hydraulic residence times or above critical organic loading rates may accumulate more VFAs than can be effectively consumed by methanogens, resulting in a low pH and consequently inhibitive effects on gas production (Ghosh, 1987).

Acidic conditions can also inhibit methanogenesis by increasing hydrogen sulfide concentrations (Metcalf and Eddy, Inc., 2003). The unionized hydrogen sulfide (H_2S) is more toxic to methanogens than the ionized form (HS^-) (Metcalf and Eddy, Inc., 2003). Acidic environments incur the unionized form of hydrogen sulfide, as described in the following equilibrium equations at a temperature of 25 °C (Metcalf and Eddy, Inc., 2003).

$$H_2S_{(g)} \leftrightarrow HS^-{}_{(aq)} + H^+ \qquad K_{a1} = [HS^-][H^+]/[H_2S] = 1\text{x}10^{-7} \text{ (mole/L)} \qquad (3)$$

$$HS^-{}_{(aq)} \leftrightarrow S_2^-{}_{(aq)} + H^+ \qquad K_{a2} = [S_2^-][H^+]/[HS^-] \approx 1\text{x}10^{-19} \text{ (mole/L)} \qquad (4)$$

Acidic biosolids are also known to have higher dissolved metal concentrations in the aqueous phase (Garber, 1982). The concentrations are typically not problematic for recycling back into the influent stream (Garber, 1982). However, metal concentrations are strictly regulated for land application (U.S. Environmental Protection Agency, 1999).

2.5b Alkalinity

Another important performance parameter is alkalinity or the acid neutralizing capacity, which is a key component to maintaining the neutral pH needed for stable digestion (Ahring, 1994; American Public Health Association, 1998). For stable digestion, total alkalinity generally ranges from 1000 to 5000 mg/L as $CaCO_3$ for the typical CO_2 content of gas produced in anaerobic digestion of municipal sludge (Grady et al., 1999; Liu and Sung, 2002). Bicarbonate (HCO_3^-) associated with the cations ammonium, calcium, or magnesium is the predominant source of buffering capacity in anaerobic digesters (Metcalf and Eddy, Inc., 2003). The importance of ammonium as a counter-ion for bicarbonate can be seen in the following equation with $C_{10}H_{19}O_3N$ representing primary solids (Grady et al., 1999; Parkin and Owen, 1986).

$$C_{10}H_{19}O_3N + 4.69\ H_2O \leftrightarrow 5.74\ CH_4 + 2.45\ CO_2 + 0.2\ C_5H_7O_2N + 0.8\ NH_4HCO_3 \qquad (5)$$

Calcium, magnesium and ammonium bicarbonate are generally proportional to the total solids concentration in the feed (Metcalf and Eddy, Inc., 2003). Figure 2.3 illustrates a correlation between higher effluent alkalinity (mg/L as $CaCO_3$) and higher total solids influent content (%) from studies on TAD.

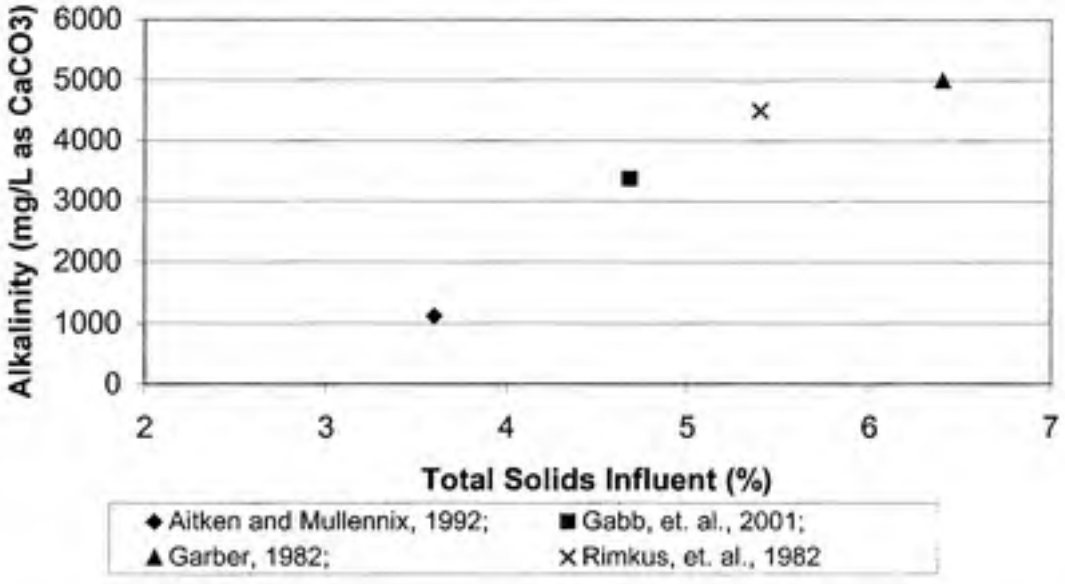


Figure 2.3. Relationship between effluent alkalinity and total solids in influent sludge in similar bench- and full-scale TAD studies with feed sludges of either primary or mixed primary and secondary (Garber, 1982; Rimkus et al., 1982; Aitken and Mullennix, 1992; Gabb et al., 2001).

Alkalinity consumption is primarily due to carbon dioxide (CO_2) and subsequent hydration to carbonic acid (H_2CO_3), as well as by VFA accumulation (Metcalf and Eddy, Inc., 2003). The bicarbonate alkalinity concentration is dependent on the pH and the partial pressure of CO_2, which is temperature-dependent according to Henry's Law (Metcalf and Eddy, Inc., 2003; Grady et al., 1999; Parkin and Owen, 1986). This relationship can be expressed in the following equation (Grady et al., 1999):

$$S_{Balk} = 6.3 \times 10^{-4} (p_{CO2}/10^{-pH}) \quad (6)$$

Where: S_{Balk} is the bicarbonate alkalinity in mg/L as $CaCO_3$

p_{CO2} is the partial pressure of carbon dioxide

Stable digestion is often associated with relatively low VFA concentrations, with bicarbonate alkalinity approximately equaling total alkalinity (Parkin and Owen, 1986). High bicarbonate alkalinity is generally indicative of low VFA concentrations, and thus associated with efficient and stable digestion. When rising VFA concentrations neutralize more buffering capacity, pH values drop and thus create further potentially inhibitive acidic conditions (Parkin and Owen, 1986). The influence of VFA concentrations on bicarbonate and total alkalinity can be also be expressed in the following equation (Parkin and Owen, 1986):

$$S_{BAlk} = T_{Alk} - 0.71(S_{VFA}) \quad (7)$$

Where: T_{Alk} is total alkalinity concentration as mg/L $CaCO_3$

S_{VFA} is volatile fatty acid concentration as mg/L as acetic acid

0.71 is the factor that converts the VFA concentration as acetic acid to $CaCO_3$ (accounting for the fact that only 85% of VFA anions are titrated to acid form at pH 4.0)

2.5c Total and Volatile Solids

Solids content is the most important physical characteristic of wastewater (Metcalf and Eddy, Inc., 2003). Solids are quantified in terms of total and volatile solids. Volatile solids are the organic or ignitable fraction of total solids, mostly consisting of organic solids although there are a few exceptions (Metcalf and Eddy, Inc., 2003). Typical ranges of solids reduction are 45 to 50% total solids, 40 to 70% volatile solids when feeding primary sludge, 20 to 50% when

feeding waste activated sludge, and 40 to 60% when feeding a combination of primary and waste activated sludge (Parkin and Owen, 1986). While solids destruction is a definitive result of stabilization, it is not as immediately responsive to changes in digester performance as pH, alkalinity, or gas production (Parkin and Owen, 1986).

Theoretically, increased operating temperatures should lead to increased biochemical conversion rates and provide increased solids destruction for a given residence time (Grady et al., 1999; Buhr and Andrews, 1977). In one of the earliest studies (in 1930), Rudolph and Heukelekian observed greater percentages of volatile solids destroyed with thermophilic batch reactors compared to a mesophilic reactor for a given retention time (Buhr and Andrews 1977). As retention time increases the disparity in solids destruction due to temperature diminishes, illustrating that provided a long enough retention time the same solids destruction will be achieved regardless of temperature (Buhr and Andrews 1977).

2.5d Total ammonium nitrogen

Total ammonium nitrogen (NH_4^+-N + NH_3-N) or TAN is present in the influent sludge and can be formed when degrading nitrogenous compounds, mainly proteins (Grady et al., 1999; Parkin and Owen, 1986). Ammonia nitrogen is an essential nutrient and considered beneficial between concentrations of 50 and 200 mg/L (Parkin and Owen, 1986). However, ammonia nitrogen can also inhibit methanogenesis with excessive concentrations of either NH_4^+ or NH_3, depending on the pH (Parkin and Owen, 1986). Free ammonia (NH_3-N) is the most toxic form and studies indicate toxic responses with concentrations around 100 mg/L (Grady et al., 1999). The concentration of free NH_3 over the range of observed pH can be estimated from the known equilibrium constant at 25 °C for the dissociation reaction $NH_4^+ \rightarrow NH_3 + H^+$ ($pK_a = -\log_{10} K_a = 9.24$; Dean, 1985), the effect of temperature on the equilibrium constant, and the measured concentration of total ammonium-nitrogen. Assuming that the enthalpy of the reaction is independent of temperature over the temperature range of interest (25 °C to 55 °C), the effect of temperature on the acid dissociation constant can be estimated from the corresponding integrated version of the van't Hoff equation (Stumm and Morgan, 1996):

$$ln\left(\frac{K_2}{K_1}\right) = \frac{\Delta H^\circ}{R}\left(\frac{1}{T_1} - \frac{1}{T_2}\right) \qquad (8)$$

Where: K_2 and K_1 = equilibrium constants for the dissociation reaction at the thermophilic temperature and at 25 °C, respectively

$\Delta H°$ = enthalpy of the dissociation reaction at the reference temperature (25 °C)

R = the universal gas constant

T_1 = the reference temperature in Kelvin

T_2 = the thermophilic temperature in Kelvin

The value of $\Delta H°$ is a function of the heats of formation (H_f) of the reactants and products at 25 °C:

$$\Delta H^\circ = H_f\left(NH_{3,aq}\right) + H_f\left(H^+\right) - H_f\left(NH_4{}^+\right) \quad (9)$$

Substituting values for the constants (Dean, 1985), the pK_a value for ammonium dissociation at 55 °C is estimated to be 8.40, compared to a value of 9.24 at 25 °C. Substituting the heats of formation constants $H_f(NH_{3,aq})$ = -19.19 kcal/mol and $H_f(NH_4{}^+)$ = -31.67 kcal/mol, the change in enthalpy, $\Delta H°$, is 12.48 kcal/mol. The fraction of the total ammonium-nitrogen concentration present as free NH_3 at pH 7.0 and 7.5 for the three temperatures studied in this project is summarized in Table 2.7. Using the actual values of average pH and average ammonium-N concentration over all of the operating conditions (Table 4.4).

Table 2.7. Estimated Fractions of Free NH_3 as a Function of pH and Temperature

	Fraction of Total Ammonium-N [a] as NH_3-N	
Temperature, °C	**pH 7.0**	**pH 7.5**
51	0.030	0.090
53	0.034	0.100
55	0.038	0.111

[a] Total $NH_4{}^+$-N + NH_3-N.

Another factor that can affect the toxicity of ammonia-nitrogen is microbial acclimation (Liu and Sung, 2002). Microbial communities can acclimate to environments with higher ammonia-nitrogen concentrations (Liu and Sung, 2002). Over time thermophilic methanogens can tolerate TAN concentrations of 3,000 mg/L and pH values as high as 8.0 (Liu and Sung,

2002). Acclimated methanogens also reach maximum methane production rates between pH values of 7.0 and 7.5 and at the NH_3-N concentrations at which acclimation occurred, with lesser or greater concentrations causing lags in gas production (Liu and Sung, 2002). Lethal total ammonia nitrogen concentrations, causing cessation of gas production regardless of acclimation, was 10,000 mg/L NH_3-N (Liu and Sung, 2002).

2.5e Volatile fatty acids

VFAs are the product of the fermentation of hydrolyzed organic compounds (Ahring, 1994; Parkin and Owen, 1986). VFA concentrations are commonly expressed in terms of the major component, acetic acid, by converting the molar concentration of each acid to the mass concentration of an equimolar concentration of acetic acid (Parkin and Owen, 1986). The range of VFA concentrations in digesting sludge is broad because they are intermediate compounds in anaerobic digestion and their accumulation depends on feed type and loading rates (Rimkus et al., 1982; Parkin and Owen, 1986). Tolerable VFA concentrations can range from 100 to 4,000 mg/L as acetic acid, depending on pH and alkalinity (Rittmann and McCarty, 2001). However, VFA concentrations around 5,500 mg/L combined with a pH of 7.0 results in concentrations of unionized volatile acids that are potentially inhibitive to methanogenesis (Parkin and Owen, 1986).

VFA concentrations are critical at thermophilic temperatures, because the increased operating temperatures increase hydrolysis proficiency while inhibiting propionate and acetate degradation and acidic conditions can exacerbate VFA accumulation (Buhr and Andrews, 1977; Garber, 1982; Ghosh, 1987). Acidic conditions are potentially troublesome because acetate-consuming methanogens are sensitive to acidic conditions and acid accumulation can escalate to a terminal state (Ghosh, 1987; Rittmann and McCarty, 2001). Increased VFA production combined with decreased consumption is why thermophilic operating temperatures often lead to 2 to 2.5 times greater VFA concentrations relative to mesophilic temperatures (Garber, 1982).

Due to the inhibitive potential of high VFA concentrations, the VFA: alkalinity ratio is a commonly used indicator of digester stability (Grady et al., 1999). A VFA: alkalinity ratio between 0.5 and 1.0 mg as acetate per mg as $CaCO_3$ is commonly associated with stable thermophilic digestion (Cabirol et al., 2002). An abnormally high VFA: alkalinity ratio caused by an accumulation of volatile acids relative to the buffering capacity can indicate a population

imbalance of the slower growing methanogens relative to the faster growing fermenters (Helmerand Hepanhol, 1997; Ahring et al., 2002). Often, thermophilic digestion results in twice the VFA: alkalinity ratio than that seen in mesophilic digestion for similar loading rates (Garber, 1982). Decreased retention times or increased loading rates, considered operating advantages to TAD, can also contribute to higher VFA concentrations (Zabranska et al., 2000; Rimkus et al., 1982; Parkin and Owen, 1986).

2.5f Gas production

The ultimate products of anaerobic digestion are methane and carbon dioxide gases, and therefore the gas production is also an important measure of stabilization (Ahring, 1994; Grady et al., 1999). Typical gas composition ranges from 65 to 70% methane (CH_4) and 25 to 30% carbon dioxide (CO_2) by volume, with small percentages of N_2, H_2, and H_2S (Zabranska et al., 2000; Metcalf and Eddy, Inc., 2003). These general gas production percentages are illustrated stoichiometrically in the following equation with $C_{10}H_{19}O_3N$ representing primary sludge (Grady et al., 1999).

$$C_{10}H_{19}O_3N + 4.69\ H_2O \rightarrow 5.74\ CH_4 + 2.45\ CO_2 + 0.2\ C_5H_7O_2N + 0.8\ NH_4HCO_3 \qquad (10)$$

The differences in composition depend on the feed. Feed sludge with higher carbohydrate concentrations produces more CO_2 compared to feed sludge with higher protein concentrations (Grady et al., 1999). Gas production is commonly normalized per unit weight of volatile solids destroyed or fed, since these are the major sources of biodegradable organic matter in sludge (Grady et al., 1999). In general, total gas production usually ranges from 0.7 to 1.12 m^3/kg VS destroyed and a common conversion for methane production is around 0.7 m^3/ kg VS destroyed (Grady et al., 1999; Metcalf and Eddy, Inc., 2003).

Methane can be expressed as oxygen demand, as can the organic content of sludge. In mass units, the COD content is 4 kg COD/kg CH_4 (Metcalf and Eddy, Inc., 2003). Assuming anaerobic conditions and standard temperature and pressure, this corresponds to 0.35 m^3 CH_4/kg COD converted (Grady et al., 1999; Rittmann and McCarty, 2001; Metcalf and Eddy, Inc., 2003). Although conflicting data exist, overall, gas yield tends to increase with temperature and more definitively increases with residence time up to an asymptotic value determined by the concentration of biodegradable volatile solids fed (Buhr and Andrews, 1977; Grady et al., 1999).

3. MATERIALS AND METHODS

3.1 Experimental Design

3.1a General approach

Experiments were designed to evaluate the CBFT[3] process for performance and pathogen inactivation capacity at different operating temperatures and with different source sludges. A lab-scale CMFR was operated continuously and periodic transfers were made from the CMFR into the CMBR to model the performance and inactivation capability of the CBFT[3] treatment concept. The lab-scale system was operated at 51, 53 and 55 °C with sludge from the South Columbus (GA) Water Resources Facility (SCWRF). In addition, the CBFT[3] process was also evaluated at 53 °C with sludges from the Orange Water and Sewer Authority (OWASA) Mason Farm Wastewater Treatment Plant in Chapel Hill, NC, and the Western Lake Superior Sanitary District (WLSSD) wastewater treatment plant in Duluth, MN. Human pathogen surrogates *Ascaris suum* and vaccine strain poliovirus were spiked into the CMFR feed sludge during all operating conditions. The CMBR was spiked during designated inactivation rate experiments. The focus of this study is confined to evaluating the physical and chemical performance parameters of the CMFR. The microbial aspects of the project, including operation of the CMBR, were beyond the scope of this report.

Conventional parameters evaluated in the feed sludge and effluent from the continuous digester included total and volatile solids (TS and VS, respectively); pH; alkalinity; VFAs, both by titration and by gas chromatography; ammonium-nitrogen; gas production; and gas composition (methane and carbon dioxide).

3.1b General operation

The primary reactor under continuous operation was a 20 L effective volume CMFR. Figure 3.1 provides a flow diagram of continuous operation. Source sludge shipments were stored and intermittently mixed in a sludge reservoir located in a large cooler. Transfers were made from the sludge reservoir to a continuously mixed feed tank located in the same cooler. Feed sludge was semi-continuously pumped from the feed tank to the CMFR located in a heated water bath. Effluent biosolids from the CMFR were pumped simultaneously with the feed pump. The effluent was stored in the effluent tank also located in the cooler. The physical layout is illustrated in more detail in the Appendix 1. A plan view is included as Figure A1.1 and two

profile views are also included as Figure A1.2 and Figure A1.3 along with a corresponding legend for the profile views, labeled Legend A1.1.

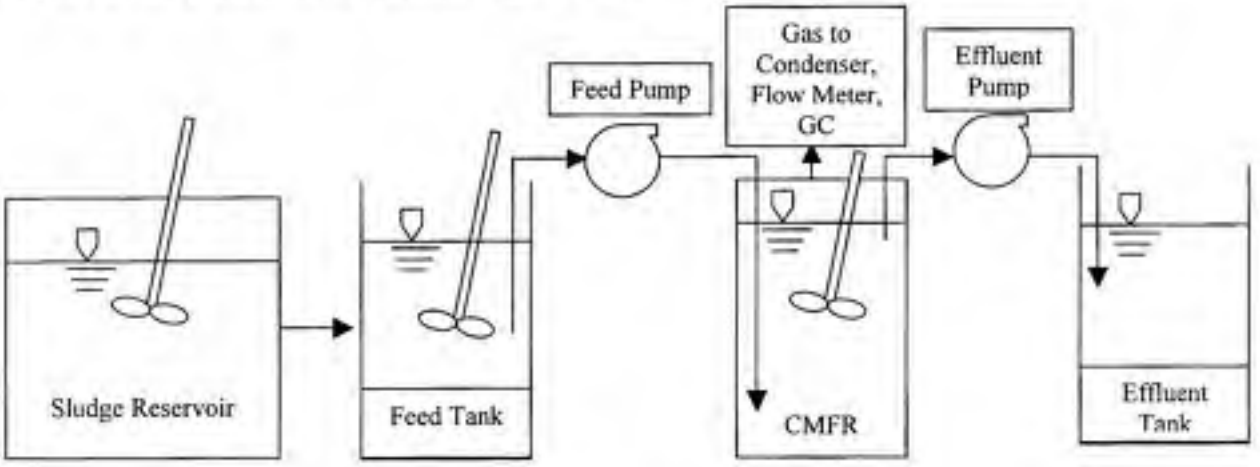


Figure 3.1. Flow diagram of continuous operation

A table documenting a general chronology of overall operation is included in Appendix 2 as Table A2.1. A table documenting the date ranges over which data were included for each operating condition is included in Appendix 2 as Table A2.2. Operation at 55 °C was conducted over two different periods (designated as periods "A" and B") because of unstable reactor performance between these periods. For each operating condition, effluent data for the physical and chemical parameters are reported for time periods after at least one residence time elapsed. The feed sludge data are reported for the entire period of operation once the target residence time was reached.

The hydraulic retention time (HRT) and the solids residence time are the same in a mixed, continuous-flow reactor without recycle, and are defined as the reactor volume divided by the average flow rate. The average flow rate was calculated using a linear fit of cumulative effluent volume vs. time. The average flow rate is illustrated in Appendix 3 as Figure A3.1. The target residence time at the beginning of the study (operation with SCWRF sludge at 55 °C) was four days. Due to concerns with reactor instability that were subsequently determined to be caused by toxic substances in the *Ascaris* and poliovirus preparations used to spike the feed sludge, the residence time was increased to between 5.3 and 6 days for all other operating conditions. These residence times are shorter than those normally used for single-stage anaerobic digestion, including thermophilic anaerobic digestion. Accordingly, the short residence times used were

meant to provide conservative data on pathogen inactivation in single-stage continuous-flow digesters.

3.2 Physical Design and Operation

3.2a Temperature control

The CMFR was suspended in the same insulated water bath maintained at constant temperature with duplicate precision constant-temperature immersion circulators (Techne TU-20D, rated temperature stability 0.005 °C; Cole-Palmer Instrument Co., Vernon Hills, IL). The constant-temperature circulators were calibrated by measuring the bath temperature with a NIST-traceable thermometer (ASTM No. 65C; Fisher Scientific, Pittsburgh, PA) readable to 0.05 °C increments.

The water bath was made up of a 125-L rectangular polyethylene tank, surrounded on all four sides and on the bottom with 1.2-cm-thick aluminum-faced rigid foam board. The top was covered with an aluminum plate with holes cut out for the reactors and the constant-temperature circulators. The water bath was refilled when necessary by pouring water through a hole in the cover, which was normally closed with a rubber stopper to prevent evaporation. Low water level was indicated on the display of the constant-temperature circulators. The water level in the bath never dropped below the sludge level in the CMFR.

The temperature in each reactor was measured with thermistors (YSI Type 401, rated accuracy ± 0.1 °C; Cole-Palmer) in direct contact with the biosolids. The thermistors were connected to precision electronic thermometers (Cole-Palmer Digi-Sense ThermoLogR, rated accuracy ± 0.03 °C) and periodically calibrated against the NIST-traceable thermometer using the water bath as the calibration medium. The thermistors were not re-calibrated if the electronic thermometer reading was within 0.05 °C of the NIST-traceable thermometer reading. The thermistors were replaced whenever the electronic thermometer readings began to drift away from the set point, which occurred only once during the period of operation for which the data are reported. Based on the rated accuracy of the thermistors and their consistency, the overall temperature accuracy of the system is considered to be ± 0.1 °C.

Tables documenting the calibration of the thermistors and constant-temperature circulators can be referenced in Appendix 4 in Table A4.1 and Table A4.2, respectively.

3.2b Sludge processing and storage

The target concentration of total solids (TS) for each sludge source, except for OWASA sludge, was 3.5% by weight, which was obtained by blending thickened and unthickened sludges. Source sludges from the SCWRF and WLSSD plants were obtained by staff at the respective plants. Source sludge from the OWASA plant was obtained by the author with the assistance of OWASA personnel.

Sludge from the SCWRF was shipped once a week in three 20-L tanks in two large coolers packed with bags of ice. Upon arrival, the carboys were stored in a cold room with a consistent temperature of 5 °C.

To process or prepare the sludge for feeding the CMFR, the sludge was transferred from the shipping carboy to a 60-L (15 gal) polyethylene drum with a marine sewage macerator pump (Jabsco model 18590-1000, ITT Industries, White Plains, NY). The macerator pump impeller served as a cutting blade to reduce the size of large solids. The drum was purged with argon prior to and during processing to minimize exposure of the sludge to air. The OWASA sludge was processed further by blending the entire volume with a submersible grinder pump (Goulds Pumps model RGS2012, 1.5 kW [2 hp], ITT Industries).

The blended sludge was pumped through a wire-mesh screen (0.6 cm openings) into a 145 L polyethylene tank referred to as the sludge reservoir, within a refrigerated cooler kept at 4 to 7 °C. The cooler was a commercial chest freezer modified by installing a temperature controller (BOD-Cubator, N-CON Systems Co. Inc., Crawford, GA) and by replacing the cover with an acrylic sheet with holes cut out for sludge and odor control tubing.

The sludge reservoir stored an approximate three to four weeks supply of feed sludge, based on the CMFR feed rate, which was thought to provide adequate equalization to mitigate changes in the feed composition. The reservoir was fitted with a detachable acrylic cover and mixed for 30 seconds every hour with a submersible sump pump (Grundfos 0.3 kW [0.4 hp] Type 316 stainless steel/polypropylene sewage ejector pump, Grundfos Pumps Corp., Olathe, KS), which was modified to discharge through a polyvinylchloride conduit with holes near each corner of the reservoir. Approximately every 3.5 days, 20 L of sludge was transferred from the reservoir to a polyethylene tank, also located in the cooler, which served as a feed tank for the CMFR. The feed tank was mixed continuously (Lightnin LabMaster SI, Lightnin Inc., Rochester, NY). A

low flow rate of argon was pumped to both the feed tank and reservoir to purge the headspace of oxygen.

The temperature of the sludge in the storage cooler was estimated by placing a thermistor probe from the feed tank mixer into a bottle of water inside the storage cooler. The ambient temperature of air in the storage cooler was monitored by hanging the probe from an indoor/outdoor digital thermometer (RadioShack Corp., Fort Worth, TX) inside the cooler.

3.2c Cleaning of sludge reservoir

In early February 2003, fungal contamination was found in samples of feed sludge during microbial analyses. To minimize fungal contamination and its potential to interfere with the microbial analyses, we removed the reservoir and the sump pump and cleaned the reservoir, its cover, and the sump pump with a chlorine bleach solution (approximately a 1:50 to 1:100 dilution of household bleach). The reservoir was cleaned again after each period of operation with a given sludge source was completed (in March, 2003 before switching to the OWASA sludge, in April, 2003 before switching to the WLSSD sludge, and in May, 2003 when switching back to the SCWRF sludge).

Cleaning consisted of rinsing the reservoir and cover with a garden hose, then filling the reservoir with the bleach solution and submerging the cover in the bleach solution. Particulate material on the interior sides of the reservoir and cover were removed with a sponge. The sump pump and dispersion conduit were cleaned by first washing the exterior surfaces and the inside of the manifold piping with a garden hose. The sump pump and conduit were then submerged in the bleach solution in the reservoir, and the pump was turned on for several minutes. After at least one hour of contact, the bleach solution was decanted, the reservoir was rinsed with the garden hose, and the reservoir was then refilled with tap water. The sump pump was rinsed with the garden hose, then submerged in the reservoir and turned on again for several minutes. The tap water was decanted from the reservoir and all surfaces were rinsed again with the garden hose.

3.2d Pumping

Feed sludge and effluent biosolids were pumped with individual peristaltic pumps for each (MasterFlex computerized drive with Easy-Load head and high performance precision Norprene

L/S 35 tubing, 7.9 mm inside diameter; Cole-Palmer). The pumps operated on timed cycles, running for one minute and remaining idle for a variable amount of time (typically from five to eight minutes). The pumps were controlled from a personal computer using software supplied by the pump manufacturer. The software allowed for adjusting the cycle times and rotation speeds for each pump, and recorded the number of cycles elapsed. Based on effluent volume measurements and elapsed cycles, the approximate volume pumped per cycle was 22 to 23 mL. Desired retention times in the CMFR were obtained by adjusting the off-time per cycle. Fatigued tubing was advanced through the pump heads periodically and replaced as necessary.

3.2e CMFR volume measurement and control

The volume of biosolids in the CMFR was measured with a floating gauge made of a hollow plastic cylinder situated inside the reactor. A thin stainless steel rod attached to the float extended through the top of the reactor into a clear plastic tube that was attached to the reactor headplate by an air-tight compression fitting. The plastic tube was labeled with one-liter increments from 15 to 20 L based on the known dimensions of the reactor. The label on the gauge was calibrated by adding an exact 20-L volume to the reactor, then using the tube height per liter to mark the remaining volumes.

At the top of the plastic tube that encased the float gauge there was a magnetic switch connected to a peristaltic pump. A magnet was attached to the top of the float gauge rod so that when the reactor volume increased to approximately 20.4 L the magnet triggered the magnetic switch and an overflow pump would turn on. The reactor volume would be decreased to approximately 19.3 L while the excess volume was pumped into the same tank used to collect reactor effluent.

3.3 Physical and Chemical Measurements

3.3a Flow rate measurement

Effluent from the CMFR was collected in a polyethylene tank located in the sludge storage cooler. Periodically the effluent volume was measured in either a 4-L or 2-L polyethylene graduated cylinder before being disposed of. The measured volumes, corresponding time, and pump cycle count were recorded and entered into a spreadsheet (Microsoft Excel, Microsoft Corp., Bellevue, WA). The average flow rate for a given operating condition was determined as

the slope of a linear regression of cumulative effluent volume vs. time. Correlation coefficients (r^2 values) were greater than 0.999 for all operating conditions. Graphs illustrating the cumulative volume vs. time for each operating condition are shown in Appendix 3 as Figure 3.1.

3.3b Gas flow

Headspace pressure in the CMFR was measured with a digital pressure gauge (Scientific Technologies Inc., model PG1000, Fremont, CA). The headspace pressure was generally maintained at 0.7 to 2 kPa (0.1 to 0.3 psi) gauge. To protect the instrumentation, water vapor was remove from the off-gas by running the off-gas line through a condenser and then to a backpressure regulator (Airtrol RV5300-3.5, McMaster Carr, Atlanta, GA) and digital gas flow meter (Omega FMA-1600, Omega Engineering Inc., Stamford, CT). The condenser consisted of a jacketed stainless steel cylinder (2.5 cm diameter, 28.5 cm long) through which chilled water (2 °C) from a constant-temperature circulator was pumped continuously. Condensate from the condenser flowed by gravity back into the CMFR.

The gas flow meter was calibrated with a bubble flow meter at three different times on two different dates (Test 1, Test 2 on 9/19/02 and Test 3 on 11/2/02). The calibration measurements and their correlation to actual gas flow rates are documented in Appendix 5 as Table A.5.1,2,3.

Upstream of the flow meter, a side stream of the off-gas from the CMFR ran continuously through a gas chromatograph (GC) sampling loop for periodic analysis by injecting automatically. The flow rate through the sampling loop was less than 2% of the total gas flow rate and was accounted for in the gas flow data.

3.3c Data acquisition

Readings from the constant-temperature circulators, electronic thermometers, and gas flow meter were sent to a data acquisition system and recorded automatically into an Excel spreadsheet on a personal computer. A general schematic of the data acquisition system is included in Appendix 1 on Figure A1.6. The data acquisition system consisted of an Edgeport/8 RS-232 to USB converter (Inside Out Networks, Austin, TX), which connected the individual sensors to the main computer; WinWedge RS-232 data acquisition software (TAL Technologies, Inc., Philadelphia, PA), which parsed and buffered the data; and Excel, which was used to log the data. Microsoft's integral Visual Basic for Applications was used to write the code that

allowed setting data capture intervals, formatting of spreadsheets, logging the data, and automated archiving of spreadsheet files.

Typically, the data were recorded at intervals of 15 to 30 minutes. Readings from the thermistors and circulators were recorded directly. However, due to the sensitivity and correspondingly rapid fluctuations of the readings from the gas flow meter, those readings were sent every six seconds to a temporary Excel spreadsheet, which was used to provide an average meter reading over the desired recording interval in the main data recording spreadsheet.

3.3d Composition of off-gas from the CMFR

Gas composition was measured with a Gow-Mac series 350 GC with thermal conductivity detection (Gow-Mac Instrument Co., Bethlehem, PA). Gas samples flowed continuously through the 1 mL sample loop and were automatically injected on intervals ranging from four to six hours. The GC was operated isothermally at 40 °C with a detector temperature of 150 °C and detector current of 150 mA. The column was a CTR1 coaxial packed column (Alltech Associates, Deerfield, IL), 2-m long x 0.6 cm diameter. Injection valve control and data acquisition, integration and analysis were done using an SRI Model 203 PeakSimple Chromatography Data System (SRI Instruments, Torrance, CA). Concentrations of methane and carbon dioxide were quantified by comparison to a two-point calibration curve prepared with standard mixtures of the two gases (80:20 v:v and 20:80 v:v, respectively; National Specialty Gases, Research Triangle Park, NC) and are reported as % by volume. Calibrations were conducted every two to three weeks. Changes between calibrations were generally very small.

Peaks for argon and nitrogen were always observed in the chromatograms but were not quantified. However, the majority of gas unaccounted for (the difference between the sum of CH_4 and CO_2 and 100%) is believed to be argon, since it was used as the purge gas. In situations when the CMFR was purged with argon, the total concentrations of CH_4 and CO_2 in subsequent samples were substantially lower than normal. Accordingly, data were rejected for any sample in which the sum of the CH_4 and CO_2 concentrations was less than 90%.

3.3e Total and volatile solids

Triplicate volumes of 25 mL each were analyzed for total and volatile solids in each feed sludge and effluent biosolids sample in accordance with method 2540G in *Standard Methods for*

the Examination of Water and Wastewater (American Public Health Association, 1998). The reported averages and standard deviations of solids measurements for a given operating period compile the averages and standard deviations for all individual measurements from all samples collected over that operating period. Solids destruction during continuous-flow treatment for a given operating condition was calculated with the following equation as illustrated for volatile solids (American Public Health Association, 1998).

$$\text{VS destroyed} = \frac{\text{VS in feed} - \text{VS in effluent}}{\text{VS in feed}} \times 100\% \qquad (11)$$

Where: VS = volatile solids concentration (g/L)

Summary statistics on the reproducibility of solids measurements are provided in Appendix 6 as Table A6.1. The average coefficient of variation (standard deviation divided by the mean) of the triplicates was less than 2% for the feed sludge and less than 1% for the effluent biosolids. The highest coefficient of variation for the feed sludge was 8% and for the effluent biosolids was 5%. No data were rejected when summarizing results for solids analyses.

Calibration records for the analytical balance used to weigh crucibles for solids analyses are documented in Appendix 7 as Table A7.1. Calibration weights were ANSI/ASTM Class 1 stainless steel (Fisher Scientific). The balance was calibrated in June 2002 and September 2002, but was not calibrated again until April 2003. At the time of re-calibration in April 2003 the error in weight relative to a typical crucible weight was less than 1% of typical changes in weight between replicate feed samples (0.8 g for total solids and 0.6 g for volatile solids) and replicate effluent biosolids samples (0.6 g for total solids and 0.4 g for volatile solids). Thus, the maximum error in weight measurements for the solids analyses was less than the standard deviation of replicate measurements for a given sample.

3.3f Preparation of samples for chemical analyses

Samples were prepared for analysis of pH, alkalinity, volatile fatty acids (VFAs), and ammonium-nitrogen (NH_4^+-N) by centrifugation and subsequent filtration as needed. All samples for chemical analysis were centrifuged at 3,000 rpm for 10 to 15 minutes. Centrifuged samples were used directly for pH and alkalinity measurements, both of which were conducted immediately after centrifugation. Centrifuged samples used for VFA and NH_4^+-N analysis were

filtered through glass fiber filters (1 μm pore size) and filtered further (0.45 μm pore size) for VFA analysis by GC.

3.3g pH

pH was measured with an Orion 710 pH meter and Orion Triode Combination pH Electrode/ATC Probe (Fisher Scientific), which provided automatic temperature compensation of the pH measurement. The meter was calibrated immediately before each use with three NIST-traceable calibration buffers (pH 4, 7 and 10; Fisher Scientific). Calibration buffers were replaced with fresh solutions weekly.

3.3h Alkalinity and VFAs by titration

Total alkalinity and alkalinity associated with bicarbonate and VFAs were measured by titration of supernatants from centrifuged samples. Approximately 20 to 30 mL of supernatant was measured in a graduated cylinder and poured into a 50 mL beaker, which was then placed on a magnetic stirrer. A sulfuric acid solution of known normality was titrated into the sample from a burette while simultaneously measuring the pH. The volume of acid required to reach pH 5.8 and the volume required to reach pH 4.3 were recorded. Total alkalinity was determined from the amount of acid required to reach pH 4.3. Bicarbonate alkalinity was determined from the amount of acid required to reach pH 5.8. The alkalinity associated with VFAs was calculated as the difference between total and bicarbonate alkalinity. Equivalents of sulfuric acid were converted to alkalinity as calcium carbonate by multiplying by 50 g CaCO3/eq. The VFA concentration as acetic acid was obtained by multiplying the equivalents of sulfuric acid for VFA-alkalinity by 60 g acetic acid/eq.

The sulfuric acid titrant used for alkalinity measurements was calibrated periodically against a newly prepared solution of sodium carbonate according to Method 2320 in *Standard Methods for the Examination of Water and Wastewater* (American Public Health Association, 1998). Calibration records for the acid titrant are shown in Appendix 8 as Table A8.1. Changes in the acid concentration were negligible (less than 1%) over long periods of time, so that frequent calibration was not necessary.

The volume of acid required to reach pH 5.8 in feed sludge samples was generally very low, so that reported bicarbonate alkalinity measurements for feed sludge is less accurate than the

other measurements. Bicarbonate and VFA alkalinity of feed sludge from OWASA are not reported because the initial pH was substantially less than 5.8.

3.3j Volatile fatty acids by gas chromatography

In most cases duplicate or triplicates of a given sample were prepared. Most of the filtered samples for analysis of individual VFAs by GC were diluted 1:1 (v:v) in oxalic acid; a few samples from early in the study were diluted with a mixture of methanol and oxalic acid. Diluted samples were stored cold until they were analyzed. The samples collected during the first operating period at 55 °C (September, 2002) were held for six months before analysis. The majority of the remaining analyses were conducted within 28 days of sample collection.

Glenn Walters, a PhD candidate in the Department of Environmental Science and Engineering at UNC-CH, analyzed the diluted samples with a packed column GC using a Hewlett Packard 5890 Series II gas chromatograph equipped with a model 7673 liquid autosampler and flame ionization detector (FID). Data acquisition and integration was performed with ChemStation software (Agilent Technologies, Palo Alto, CA). The carrier gas was helium set to 290 kPa (42 psi) gauge at 150 °C and the instrument's integral electronic pressure control maintained carrier gas velocity at 75 cm/s. The packed column was a 3.2-mm diameter, 2-m long stainless steel column packed with 4% Carbowax 20M on 80/120 Carbopack BDA (Supelco, Bellefonte, PA). Sample injection was splitless using the standard linerless injection port set at 200 °C with injection volume of either 2 µL or 3 µL depending on the expected concentration in the samples. The column oven temperature program began with two minutes at 150 °C followed by an increase of 4 °C/min to a final temperature of 210 °C, which was then held for five minutes. The FID temperature was set at 260 °C with nitrogen used for make-up gas. Column conditioning was performed at instrument start-up (prior to running samples) by repeated injection of 30 mM oxalic acid until the resulting unretained peak height remained unchanged between successive injections.

Individual VFAs were quantified by comparison to three-point calibration curves generated from external standards of acetic, propionic, butyric, isobutyric, valeric, isovaleric, 4-methylvaleric and hexanoic acids. Compounds used as standards were ACS reagent grade or the highest grade available. External standards were prepared gravimetrically and diluted to a range of concentrations that bracketed the expected concentrations of the samples. Recalibration was

typically performed approximately once every three hours during any given run. Average values of the correlation coefficients (r^2 values) for the calibration curves were 0.996 or greater for all of the VFAs.

In some cases one of the replicate analyses for a given sample was very inconsistent with the other value(s) and with the general trend in concentration for the period of operation during which the sample was collected. In such cases that value was rejected and not included in the data summary. Reported means and standard deviations of VFA concentrations for a given operating period represent the means and standard deviations of pooled data for all (non-rejected) individual measurements from all samples collected over that operating period. The molar concentrations of individual acids were summed and then converted to mg/L as acetic acid by multiplying the total molar concentration by the molecular weight of acetic acid (60 g/mol).

3.3j Ammonium-nitrogen

Ammonium-nitrogen was analyzed by the Hach Nessler Method (Hach Company, 1997) using Hach reagents (Hach Co., Loveland, CO). Each filtered sample of feed sludge or effluent biosolids was diluted 1:500 (v:v) in reagent water containing enough sulfuric acid to achieve a final pH of 2 or less. Volumetric glassware was used to make the dilutions. A method blank was prepared at the same time by replacing the sample volume with reagent water but otherwise following the same procedures as for the samples. Diluted samples and method blanks were placed in glass storage bottles and stored cold. All samples and blanks were analyzed within 28 days of collection.

When ready for analysis, samples and method blanks were neutralized with a combination of 5 N sodium hydroxide and a solution of K_2HPO_4 to bring the pH to 6 or greater. Each sample or method blank was analyzed in three 25-mL volumes. Each volume was prepared by mixing in the appropriate amounts of the Nessler reagents and was then analyzed immediately with a UV-visible spectrophotometer (Hitachi U-2000, Hitachi High Technologies America, Schaumburg, IL) at 425 nm. The spectrophotometer was zeroed with reagent water before and after all standards, samples, and method blanks were analyzed.

The concentration of NH_4^+-N in each replicate of the diluted samples and method blanks was calculated using a four-point calibration curve prepared from standards of known concentration. Standards consisted of reagent water for the zero concentration (analytical blank) and solutions

of reagent-grade ammonium chloride which was prepared gravimetrically. The ammonium chloride standards were prepared at concentrations of 0.5, 1.5 and 2.5 mg N/L. Each standard was analyzed in triplicates using the same procedure as with the samples. All calibration curves had r^2 values of at least 0.997, with all but one value at least 0.999.

All of the method blanks had optical absorbance values near or less than the value for the analytical blank, indicating that there was no detectable ammonium in the method blanks. The average coefficient of variation of the triplicates for feed samples was 2.6% and for the effluent samples was 1.5%. No data were rejected when summarizing results of the NH_4^+-N analyses. Reported averages and standard deviations of NH_4^+-N concentrations for a given operating period represent the means and standard deviations of all individual measurements from all samples collected over that operating period.

3.3k Statistical analyses of data

Means and standard deviations of data sets were calculated with Excel. Standard deviations of calculated values were determined with propagation of error formulas from the means and standard deviations of the individual values (Young, 1962). Comparisons between averages were made with *t*-tests for independent samples using TK Solver 4.0 (Universal Technical Systems, Rockford, IL). Differences between averages are considered significant if $p \leq 0.05$. Linear regressions were performed either with Grapher (Golden Software Inc., Golden, CO) or with Excel.

4. RESULTS, DISCUSSION, AND CONCLUSIONS

4.1 Results and Discussion

4.1a Operation and control

The mean temperature and residence time for the CMFR during each operating period are summarized in Table 4.2. The overall accuracy of the temperature for the CMFR and CMBR is considered to be ± 0.1 °C. The consistency of the temperature in the CMFR during a given operating period, as indicated by the standard deviation, was excellent.

Differences in the residence times for the operating periods other than those at 55 °C were due mostly to slight differences in the volume of feed sludge pumped per cycle for the different sludge sources.

4.1b CMFR feed sludge characteristics

The three sources of feed sludge were South Columbus (Georgia) Water Resources Facility (SCWRF), the Orange Water and Sewer Authority (OWASA) Mason Farm Wastewater Treatment Plant in Chapel Hill, NC, and the Western Lake Superior Sanitary District (WLSSD) wastewater treatment plant in Duluth, MN. A summary of sludge characteristics from these wastewater treatment plants is included in Table 4.1.

Table 4.1. Characteristics of Sludge Sources Used in the Project

Source	Sludge Type	Average Plant Wastewater Flow (mgd)	Approximate Contribution from Industries (%)	Primary Industrial Activity
SCWRF	primary + waste activated [a]	28	20	textile, bakery
OWASA	fermented primary	9	< 2	university
WLSSD	waste activated [b]	40	50	pulp and paper

[a] Approximately 55% primary, 45% waste activated (v:v).

[b] From a pure-oxygen plant with no primary treatment.

Table 4.2. Operating Parameters for the CMFR for All Operating Conditions [a]

Parameter	Value Over Indicated Operating Condition [b]					
	SCWRF-55A	**SCWRF-55B**	**SCWRF-53**	**SCWRF-51**	**OWASA-53**	**WLSSD-53**
Temperature, °C	55.06 ± 0.04 (556)	55.00 ± 0.02 (467)	52.98 ± 0.04 (3,061)	50.95 ± 0.04 (2,368)	52.95 ± 0.05 (2,095)	52.97 ± 0.03 (2,131)
Flow, L/d	5.00 (12)	4.56 (5)	3.46 (12)	3.50 (7)	3.33 (5)	3.80 (5)
Digester volume, L	20.1 ± 0.08 (12)	19.9 ± 0.17 (5)	20.0 ± 0.28 (9)	20.1 ± 0.16 (7)	19.9 ± 0.2 (6)	20.1 ± 0.2 (5)
HRT, days	4.02 ± 0.02	4.37 ± 0.04	5.79 ± 0.08	5.75 ± 0.04	5.98 ± 0.05	5.28 ± 0.06

[a] Operating conditions are identified by the sludge source and the operating temperature; the two periods of operation at 55 °C are designated "A" and "B" in chronological order.

[b] Data represent means ± standard deviations. The number in parentheses is the number of samples analyzed. All data are rounded to three significant figures. Units for alkalinity are mg/L as $CaCO_3$; units for VFAs as acetic acid and NH_4^+-N are mg/L.

A more extensive summary of the physical/chemical characteristics of the feed sludges is included below on Table 4.3. Differences in average solids concentrations among the sludges resulted more from the removal of solid material by screening the sludge when it was placed into the storage reservoir than from differences in the sludges as received. As expected, the OWASA sludge had a substantially lower pH than the other sludges, since it was obtained from a full-scale reactor used to ferment primary sludge. Since the OWASA sludge did not contain any waste activated sludge, the ammonium concentration was also lower than in the other sludges. Primary sludge usually contains less nitrogen per unit solids than waste activated sludge, therefore less nitrogen would be released upon digestion (Metcalf and Eddy, Inc., 2003).

The ammonium concentration in the WLSSD sludge was also low relative to the concentration in the SCWRF sludge, although the concentration increased with storage time (not shown). Therefore, differences between the WLSSD sludge and the SCWRF sludge may be due in part to the longer average storage time of the SCWRF sludge in the sludge storage reservoir.

As expected, most of the alkalinity in the feed sludges was attributable to VFAs. Differences in VFA concentration for the SCWRF sludge across the different operating periods may be related to differences in sludge characteristics at the SCWRF plant over time, or to changes during storage. The concentration of VFAs in the OWASA sludge was surprisingly low considering that the sludge was obtained from a fermentation process. Concentrations of total VFAs measured by gas chromatographic analysis of individual acids were substantially greater than concentrations measured by titration for most of the sludge samples. The titration measurement is considered to be less accurate because of the small sample volumes and correspondingly small volumes of acid titrant used. A comparison of the measurements for individual samples is shown in Appendix 9 on Figure A9.1.

Speciation of the VFAs in the feed sludge samples is shown below in Figure 4.1. Acetic and propionic acids were the most abundant acids in all of the sludges. The concentration of acetic acid was at least twice that of propionic acid in the SCWRF sludge, whereas the concentrations of the two acids were approximately equal in the OWASA and WLSSD sludges. Butyric acid was the next most abundant acid for all sludges except for the SCWRF sludge used while the CMFR was operated at 55 °C; in that case, isovaleric acid was the next most abundant acid. All other acid concentrations were 5% or less of the total VFAs.

Table 4.3. Feed Sludge Characteristics for All Operating Conditions [a]

Parameter	Value Over Indicated Operating Condition					
	SCWRF-55A	**SCWRF-55B**	**SCWRF-53**	**SCWRF-51**	**OWASA-53**	**WLSSD-53**
Solids						
total, % (wt:wt)	3.14 ± 0.12 (5)	2.99 ± 0.13 (3)	2.72 ± 0.24 (7)	3.05 ± 0.15 (5)	2.72 ± 0.35 (3)	3.48 ± 0.15 (4)
volatile, % (wt:wt)	2.36 ± 0.08 (5)	2.22 ± 0.10 (3)	2.04 ± 0.19 (7)	2.13 ± 0.11 (5)	2.05 ± 0.14 (3)	2.70 ± 0.12 (4)
volatile/total, %	75.4 ± 0.4 (5)	74.2 ± 0.6 (3)	75.1 ± 1.2 (7)	69.8 ± 1.5 (5)	76.0 ± 7.0 (3)	77.5 ± 0.4 (4)
pH	6.42 ± 0.12 (4)	6.26 ± 0.19 (3)	6.21 ± 0.20 (7)	6.15 ± 0.15 (6)	5.01 ± 0.06 (4)	6.83 ± 0.42 (5)
Alkalinity						
total	942 ± 194 (4)	1,300 ± 341 (3)	1,430 ± 448 (7)	827 ± 308 (4)	ND [c]	1,110 ± 457 (4)
bicarbonate	67 ± 22 (4)	93 ± 49 (3)	80 ± 75 (7)	52 ± 1 (4)	ND	313 ± 213 (4)
VFA	875 ± 207 (4)	1,210 ± 300 (3)	1,350 ± 449 (7)	774 ± 307 (4)	ND	795 ± 367 (4)
VFAs as acetic acid						
by titration	1,050 ± 249 (4)	1,450 ± 360 (3)	1,620 ± 539 (7)	674 ± 487 (6)	ND	954 ± 441 (4)
by GC	2,250 ± 191 (5)	ND	2,660 ± 631 (6)	1,470 ± 493 (5)	1,110 ± 250 (3)	817 ± 286 (5)
NH_4^+-N	422 ± 40 (5)	526 ± 21 (2)	625 ± 46 (7)	276 ± 76 (5)	85 ± 5 (3)	135 ± 113 (4)

[a] Notes are as in Table 4.2.

[c] ND = not determined

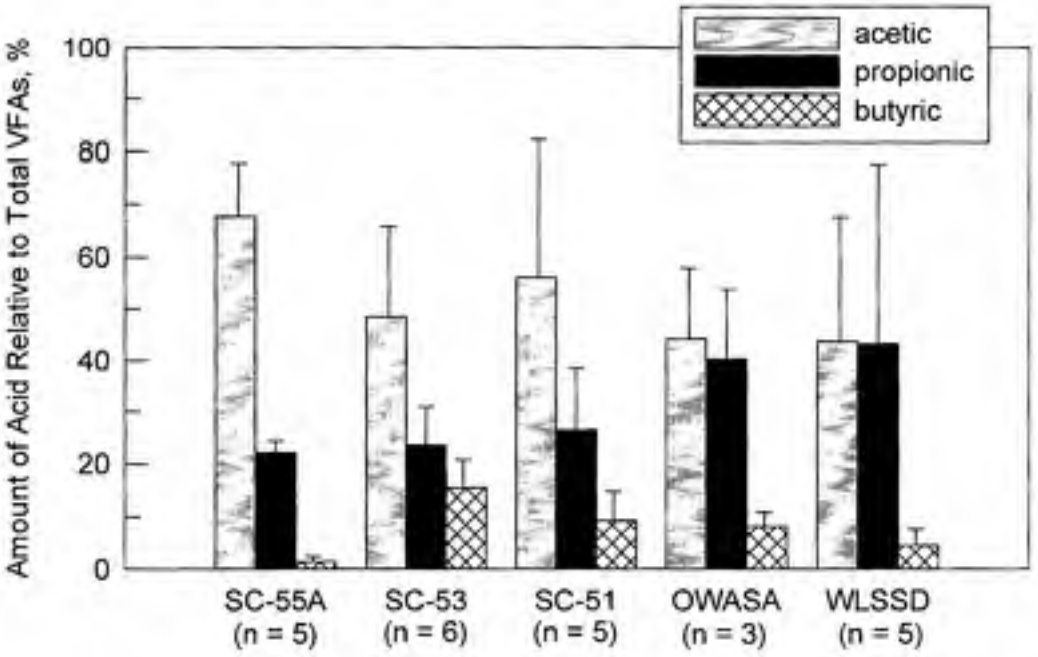


Figure 4.1. Speciation of VFAs in feed sludges for each operating condition (SC = South Columbus). Molar concentrations of each acid were used to calculate its fraction of the total VFAs, and only the three most abundant acids are shown. Error bars represent one standard deviation. The number of samples analyzed is shown in parentheses beneath the label for each operating condition.

4.1c CMFR effluent biosolids characteristics

Effluent concentrations of total and volatile solids, alkalinity, VFAs, and NH_4^+-N, as well as effluent pH, are summarized in Table 4.4 for each operating condition. Indicators of performance with respect to solids conversion and stabilization – total and volatile solids destruction and gas production per unit volatile solids fed or destroyed – are compared in Table 4.4 as well. It should be kept in mind that the residence times for operation of the CMFR at 55 °C were lower than the residence times used for the other operating conditions.

Although temperature is believed to be the primary factor responsible for the inactivation of pathogens and indicator organisms in high-temperature processes such as TAD, effluent characteristics that may also influence the inactivation of these organisms include pH, concentrations of protonated (non-ionized) organic acids, and free NH_3 (non-ionized ammonia). The removal of any pathogen or indicator organism across the CMFR is reflective of the conditions in the digester. For a CMFR, the effluent concentrations of potentially relevant parameters reflect the conditions to which the organisms were exposed in the reactor.

Table 4.4. Effluent Concentrations and Performance Parameters for the CMFR for All Operating Conditions [a]

Parameter	Value Over Indicated Operating Condition					
	SCWRF-55A	SCWRF-55B	SCWRF-53	SCWRF-51	OWASA-53	WLSSD-53
Effluent Solids						
total, % (wt:wt)	2.42 ± 0.05 (4)	2.19 ± 0.02 (2)	1.92 ± 0.05 (6)	2.16 ± 0.11 (4)	1.68 ± 0.06 (3)	2.46 ± 0.07 (3)
volatile, % (wt:wt)	1.62 ± 0.06 (4)	1.42 ± 0.02 (2)	1.26 ± 0.03 (6)	1.28 ± 0.05 (4)	1.14 ± 0.07 (3)	1.70 ± 0.05 (3)
volatile/total, %	66.6 ± 1.3 (4)	65.0 ± 0.6 (2)	65.4 ± 0.6 (6)	59.2 ± 2.3 (4)	67.5 ± 1.6 (3)	69.3 ± 1.2 (3)
Solids Destruction						
total, %	22.9 ± 1.0	26.7 ± 1.2	29.5 ± 2.7	29.4 ± 4.1	38.1 ± 5.0	29.4 ± 1.5
volatile, %	31.6 ± 1.7	35.8 ± 1.7	38.6 ± 3.7	40.2 ± 2.5	44.5 ± 4.0	37.0 ± 1.9
Effluent pH	7.38 ± 0.06 (6)	7.37 ± 0.04 (2)	7.34 ± 0.06 (6)	7.52 ± 0.07 (5)	7.02 ± 0.22 (3)	7.44 ± 0.30 (5)
Effluent Alkalinity						
total	2,840 ± 163 (6)	2,950 ± 156 (2)	3,380 ± 334 (6)	3,210 ± 101 (5)	1,760 ± 388 (3)	3,790 ± 1,080 (5)
bicarbonate	1,500 ± 149 (6)	1,550 ± 398 (2)	1,770 ± 96 (6)	2,180 ± 88 (5)	905 ± 331 (3)	2,440 ± 1,080 (5)
VFA	1,340 ± 59 (6)	1,390 ± 242 (2)	1,610 ± 270 (6)	1,030 ± 70 (5)	854 ± 62 (3)	1,350 ± 92 (5)
VFAs as acetic acid						
By titration	1,610 ± 71 (6)	1,670 ± 290 (2)	1,940 ± 326 (6)	1,230 ± 84 (5)	1,020 ± 75 (3)	1,620 ± 110 (5)
By GC	1,610 ± 204 (4)	1,460 ± 208 (2)	1,800 ± 181 (4)	672 ± 188 (4)	887 ± 106 (3)	936 ± 370 (2)
Effluent NH_4^+-N	912 ± 42 (4)	958 ± 25 (2)	1,040 ± 58 (6)	975 ± 11 (4)	557 ± 97 (3)	1,160 ± 324 (4)
Gas flow, mL/min	32.4 ± 4.2 (428)	26.8 ± 3.2 (607)	24.9 ± 2.1 (2,217)	21.3 ± 3.6 (1,487)	19.5 ± 2.9 (1,125)	22.1 ± 3.0 (1,120)
Gas production						
m^3/kg VS fed	0.40 ± 0.05	0.38 ± 0.05	0.51 ± 0.06	0.41 ± 0.07	0.41 ± 0.07	0.31 ± 0.04
m^3/kg VS destroyed	1.25 ± 0.16	1.06 ± 0.19	1.31 ± 0.34	1.02 ± 0.22	0.92 ± 0.21	0.84 ± 0.16
Gas composition						
CH_4, % (v:v)	ND	60.4 ± 1.1 (18)	62.2 ± 1.3 (70)	61.2 ± 1.2 (78)	56.7 ± 1.6 (47)	59.6 ± 2.3 (77)
CO_2, % (v:v)	ND	36.4 ± 0.4 (18)	32.4 ± 0.7 (70)	33.8 ± 1.6 (78)	36.7 ± 0.8 (47)	32.4 ± 3.0 (77)

[a] Notes are as in Table 4.3.

Depending on the method used to measure the effluent VFA concentrations, these concentrations were in the range of 1,000 to 2,000 mg/L as acetic acid. Concentrations measured by GC were generally lower than those measured by titration; a comparison of the methods for individual samples is shown in Appendix 9 in Figure A9.1. Speciation of the most abundant VFAs is shown in Figure 4.1. In contrast to the speciation in feed sludge, the major VFA in effluent biosolids was propionic acid for all operating conditions. This observation is consistent with reports by others that propionate is relatively poorly removed under thermophilic anaerobic conditions (Griffin et al., 1998; Ahring et al., 2001; Kim and Speece, 2002). Also in contrast with the feed sludges, isobutyric acid was generally the next most abundant acid after propionic and acetic acids. For the operating period with sludge from the WLSSD, valeric acid was more abundant than isobutyric acid in the effluent biosolids. For the second period of operation with sludge from the SCWRF at 55 °C, both valeric and isovaleric acids were more abundant than isobutyric acid.

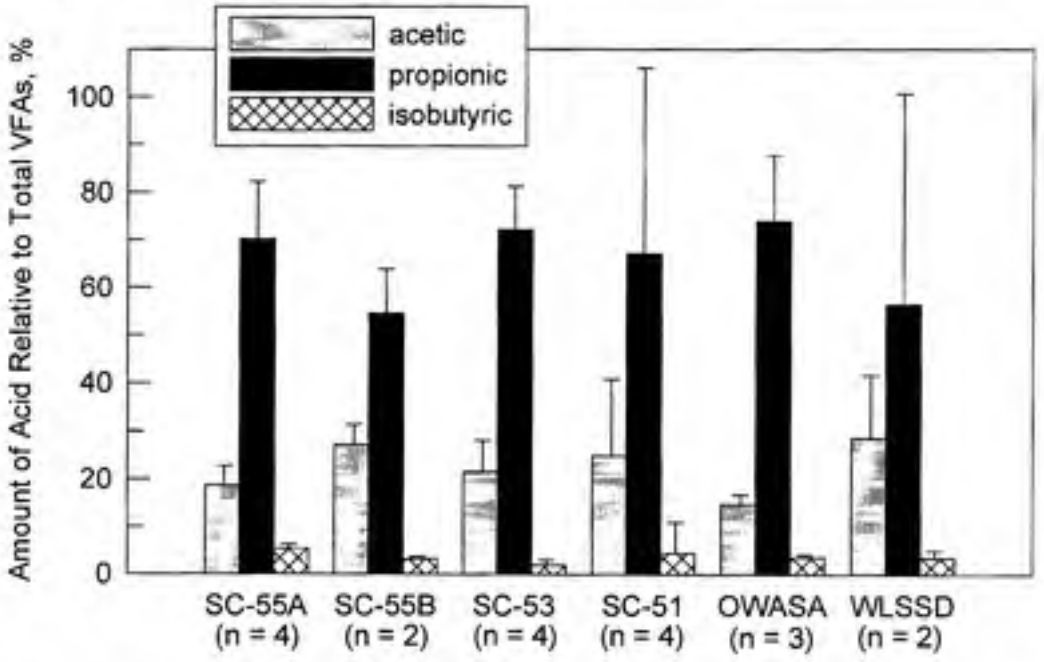


Figure 4.2. Speciation of VFAs in effluent biosolids from the CMFR for each operating condition. Notes are as in Figure 4.1.

Concentrations of NH_4^+-N in the effluent biosolids were in the 1,000 mg/L range under all conditions except during operation with OWASA sludge, for which the effluent NH_4^+-N concentration was 560 mg/L. The lower concentration during treatment with the OWASA

sludge is consistent with a low concentration of NH_4^+-N in the OWASA feed sludge and with the absence of waste activated sludge in the OWASA feed sludge.

The effluent pH was 7.0 or above for all operating conditions. The effluent pH was lowest (7.0) for the period of operation with OWASA sludge and ranged from 7.3 to 7.5 for all other operating conditions. At these pH values, the concentrations of protonated VFAs would be expected to be negligible; acid dissociation constants (pK_a values) for VFAs at 25 °C are near 4.7 (Dean, 1985). The pH values resulted in the average concentration of free NH_3 in the CMFR to range from 20 mg/L as N (for the OWASA sludge treated at 53 °C) to 100 mg/L as N (for the SCWRF sludge treated at 51 °C and the WLSSD sludge treated at 53 °C). The maximum free ammonia concentration is below the 200 mg/L NH_3 concentration that can result in the inhibition of methanogenesis (Parkin and Owen, 1986).

4.1d Solids conversion and stabilization in the CMFR

The lowest destructions of volatile solids were observed during operation at 55 °C, which also corresponded to the lowest residence time evaluated. The highest destruction of VS was observed with OWASA sludge, which was significantly higher than VS removal for both other sludges evaluated at 53 °C. This difference may reflect slightly better destruction of volatile solids in primary sludge than in waste activated sludge during TAD (see Chapter 2). Removals of VS under most operating conditions were near or below 38%, which is the target level of VS destruction required for vector attraction reduction under Option 1 of the Part 503 regulations (U.S. Environmental Protection Agency, 1999). Figure 4.3 illustrates the volatile solids destruction for each operating condition and the 38% vector attraction limit.

Gas production during the periods of operation with sludge from the SCWRF ranged from 1.0 to 1.3 m^3/kg VS destroyed. There was no apparent trend with temperature in gas production for the SCWRF sludge. Slightly less gas was produced during treatment of OWASA sludge and sludge from the WLSSD, although differences in gas production relative to that from other sludges were significant in only a few cases. Gas production from both the OWASA sludge and the WLSSD sludge was significantly lower than gas production from the SCWRF sludge during

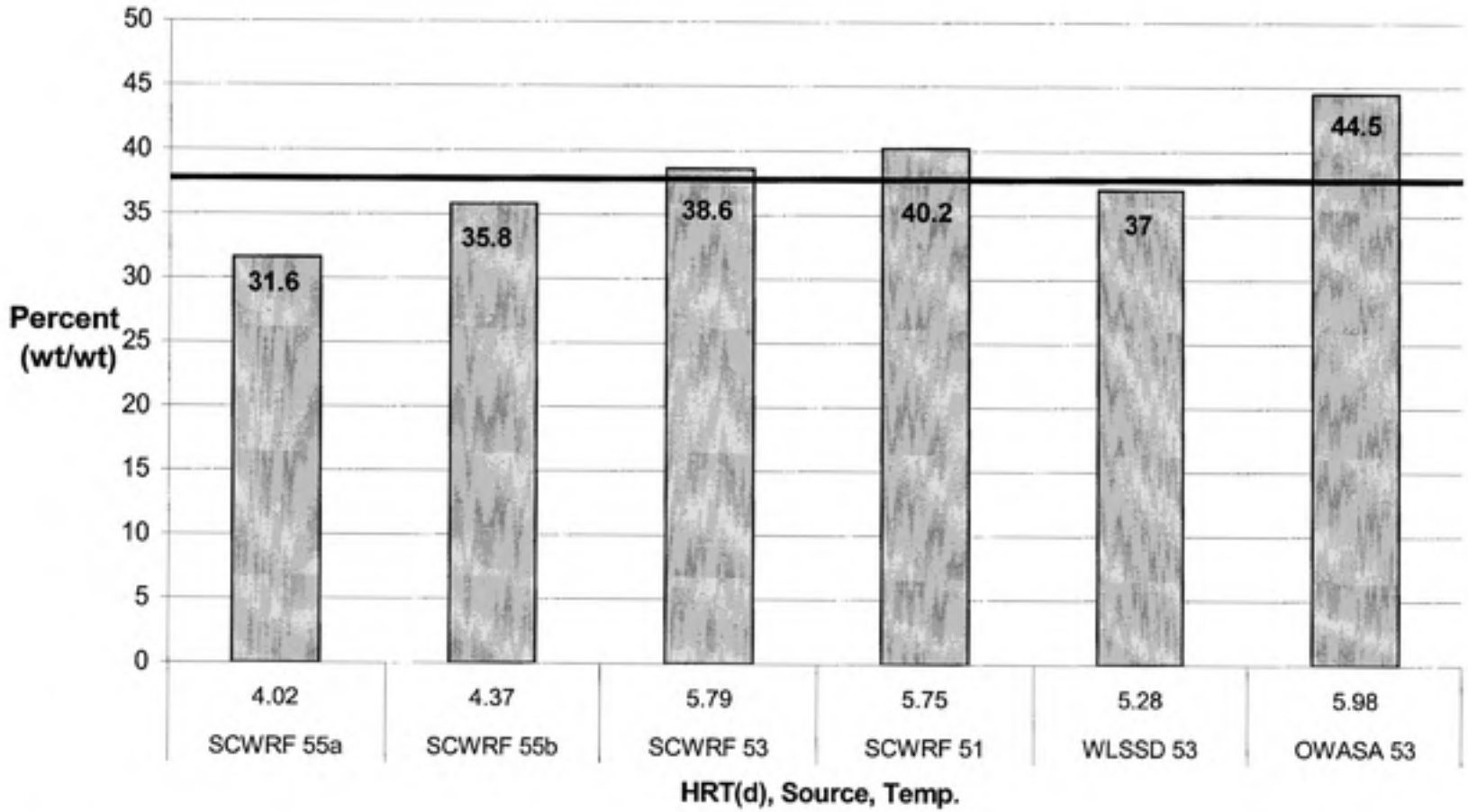


Figure 4.3. Volatile solids destruction from the CMFR for each operating condition, the bold line represents 38% VS destruction regulatory requirement for Class A biosolids.

the first period of operation at 55 °C. Gas production from the WLSSD sludge was also significantly lower than gas production from the SCWRF sludge during operation at 53 °C. All gas production values were within or slightly higher than the expected range for anaerobic digestion of wastewater sludge (Metcalf and Eddy, Inc., 2003).

4.2 Conclusions

A laboratory-scale, continuous-flow thermophilic anaerobic digester was capable of stable operation at a short residence time (four to six days) over a temperature range from 51 °C to 55 °C with influent sludge from three different sources. Except for the period of treatment with OWASA sludge, reductions of volatile solids were near or below the level required for vector attraction reduction under Option 1 of the Class A biosolids criteria in the Part 503 regulations. Although other options can be used to meet the requirements for vector attraction reduction, residence times on the order of six days may represent a lower limit for achieving the vector attraction reduction requirements under Option 1 in a single-stage thermophilic anaerobic digester. Residence times in the range used in this study might be more applicable for continuous-flow thermophilic anaerobic digestion in two-stage processes, such as the temperature-phased anaerobic digestion (TPAD) process.

Table 4.6 offers a comparison of resulting stabilization efficiency from my study and similar continuous, single-stage TAD studies. The table lists previous studies in increasing order of residence time. The table illustrates the direct relationship between increasing residence time and increasing volatile solids destruction. Volatile solids destruction values that are incongruent with this trend seem to coincide with differences in feed sludge types. The stabilization efficiency results from this study are in accordance with the findings of similar continuous, single-stage TAD studies.

Gas production from the CMFR per unit volatile solids destroyed was consistent with values typical of laboratory- and full-scale anaerobic digestion, as illustrated in Table 4.6. The pH in the digester ranged from 7.0 to 7.5. Over this pH range, it is likely that the concentrations of protonated VFAs in the digester were negligible. The most predominant VFA in the CMFR effluent was propionate, consistent with observations by others that this acid tends to accumulate

Table 4.5. Comparison of stabilization between this study and other continuous single-stage TAD studies

Reference	Scale	Feed Type[a]	Residence Time (day)	Temperature (°C)	Volatile Solids Destruction (%)	Gas Production per kg Volatile Solids	
						Fed (m^3/kg)	Destroyed (m^3/kg)
(Ghosh, 1987)	Lab	P+WAS	3.0	55	28.8	0.18	0.63
SCWRF (this study)	Lab	P+WAS	4.02	55 A[d]	31.6	0.40	1.25
SCWRF (this study)	Lab	P+WAS	4.04	55 B[d]	35.8	0.38	1.06
WLSSD (this study)	Lab	WAS	5.28	53	37.0	0.31	0.84
SCWRF (this study)	Lab	P+WAS	5.75	51	40.2	0.41	1.02
SCWRF (this study)	Lab	P+WAS	5.79	53	38.6	0.51	1.31
OWASA (this study)	Lab	P	5.98	53	44.5	0.41	0.62
(Ghosh, 1987)	Lab	P+WAS	7.0	55	59.7	0.37	0.62
(Aitken and Mullennix, 1992)	Lab	P+WAS[b]	10.0	55	43.6	0.39	0.89
(Rimkus et al., 1982)	Full	P+WAS	11.3	54	34.0	0.40	1.20
(Cabirol et al., 2002)	Lab	P+WAS	14.0	55	63.0	0.14	0.22
(Ghosh, 1987)	Lab	P+WAS	15.0	55	67.9	0.23	0.33
(Gabb et al., 2001)	Lab	P[b]	16.1	50	49.3	0.57	1.16
(Garber, 1982)	Full	P+WAS	20.0	49	65.0	0.75	1.20
(Cabirol et al., 2002)	Lab	P+WAS	20.0	55	74.0	0.07	0.10
(Cabirol et al., 2002)	Lab	P+WAS[c]	20.0	55	58.0	0.04	0.08
(Krugel et al., 1998)	Full	P+WAS	21.0	55	60.0	0.60	1.00
(Krugel et al., 1998)	Full	P+WAS	44.0	56	80.0	0.52	0.65
(Cabirol et al., 2002)	Lab	P+WAS[c]	49.0	55	33.0	0.20	0.61

[a] P is primary sludge. WAS is waste activated sludge.; [b] 40% fermented primary mixed with 60% thickened waste activated sludge; [c] coagulated primary sludge mixed with waste activated sludge; [d] A and B indicate with the first and second trials at 55 °C, respectively.

under thermophilic anaerobic conditions. The concentration of free ammonia (NH_3) in the digester was estimated to be from 20 to 100 mg/L over the range of conditions evaluated.

Future research should focus on combining batch treatment with other reactor configurations as well as modeling the $CBFT^3$ under different feeding methods and temperatures. For example, the $CBFT^3$ concept of combining batch treatment with other treatment processes could be applied beyond the single-stage continuous type of reactor to other types such as temperature phased anaerobic reactors or acid- and gas-phased anaerobic reactors. Phased anaerobic digestion has been demonstrated to achieve higher rates of volatile solids destruction and increased gas production (Ghosh, 2001). Increasing rates of volatile solids destruction would decrease total treatment retention times. Further testing the of $CBFT^3$ concept under different feeding methods such as a draw and fill feeding method would also broaden the knowledge base for extrapolating lab-scale data to more common full-scale operations.

5. REFERENCES

Ahring, B.K. (1994) Status on science and application of thermophilic anaerobic digestion. *Water Science and Technology*, **30:** 241-249.

Ahring, B.K., A.A. Ibrahim and Z. Mladenovska. (2001) Effect of temperature increase from 55 to 65°C on performance and microbial population dynamics of an anaerobic reactor treating cattle manure. *Water Research*, **35:** 2446-2452.

Ahring, B.K., Z. Mladenovska, R. Iranpour and P. Westermann (2002) State of the art and future perspectives of thermophilic anaerobic digestion. *Water Science and Technology*, **45:** 293-298.

Aitken, M.D. and R.W. Mullennix (1992) Another Look at Thermophilic Anaerobic Digestion of Wastewater Sludge. *Water Environmental Research*, **64:** 915-919.

Aitken, M.D., M.D. Sobsey, G.W. Walters, M. Shehee, P.L. Crunk, K.E. Blauth, V.R. Hill, and S.P. Napier (2003) Evaluation of Pathogen and Indicator Organisms Removal During Continuous and Batch Thermophilic Anaerobic Digestion of Wastewater Sludge. Unpublished report submitted to Brown and Caldwell, Inc., Atlanta, GA.

American Public Health Association, (1998) *Standard Methods for the Examination of Water and Wastewater, 20th Edition*. American Public Health Association, American Water Works Association, and Water Environment Federation: Washington, DC.

Buhr, H.O. and J.F. Andrews (1977) The thermophilic anaerobic digestion process. *Water Research*, **11:** 129-143.

Cabirol, N., M. Rojas Oropeza and A. Noyola. (2002) Removal of helminth eggs and fecal coliforms by anaerobic thermophilic sludge digestion. *Water Science and Technology*, **45:** 269-274.

Dean, J.A., ed. (1985) *Lange's Handbook of Chemistry*, McGraw-Hill: New York.

Gabb, D. M., J.M. Hake, C. De Leon, D. Jenkins, R.E. Danielson and S. Ghosh (2001) Comparison of thermophilic anaerobic digeston processes for meeting biosolids Class A pathogen standards. *Water Environment Federation 15th Annual Conference Proceedings*, 293-298.

Garber, W.F. (1982) Operating experience with thermophilic anaerobic digestion. *Journal of Water Pollution Control Federeration*, **54:** 1170-1175.

Ghosh, S. (1987) Improved sludge gasification by two-phase anaerobic digestion. *Jornal of Environmental Engineering*, **113:** 1265-1285.

Grady, C.P.L. Jr., G.T. Daigger and H.C. Lim (1999) *Biological Wastewater Treatment 2nd ed.* Marcel Dekker, Inc.: New York.

Griffin, M.E., D.D. McMahon, R.I. Mackie and L. Raskin (1998) Methanogenic population dynamics during start-up of anaerobic digesters treating municipal solid waste and biosolids. *Biotechnology and Bioengineering*, **57:** 342-355.

Helmer, R. and I. Hepanhol, ed. (1997) *Water Pollution Control: A Guide to the Use of Water Quality Management Principles*, UNESCO: London.

Iranpour, R., F. Alatriste-Mondragon, S.V. Diaz-Perez and H.H.J. Cox. (2003) Discussion of: The role of molecular methods in evaluating biological treatment processes by B. E. Ritmann, *Water Environment Research,* **75:** 283-284.

Kim, M. and R.E. Speece (2002) Aerobic waste activated sludge (WAS) for start-up seed of mesophilic and thermophilic anaerobic digestion. *Water Research*, **36:** 3860-3866.

Krugel, S., L. Nemeth and C. Peddie (1998) Extending thermophilic anaerobic digestion for producing Class A biosolids at the Greater Vancouver Regional Districts Annacis Island Wastewater Treatment Plant, *Water Science and Technology*, **38:** 409-416.

Liu, T., and S. Sung (2002) Ammonia inhibition on thermophilic acetoclastic methanogens. *Water Science and Technology,* **45:** 113-120.

Metcalf & Eddy, Inc. (2003) *Wastewater Engineering Treatment and Reuse, 4th Ed.*. Prentice Hall: Upper Saddle, NJ.

National Research Council Committee on Toxicants and Pathogens in Biosolids Applied to Land (2002) *Biosolids Applied to Land: Advancing Standards and Practices.* The National Academies Press: Washington DC.

Nielsen, B. and G. Petersen (2000) Thermophilic anaerobic digestion and pasteurisation. Practical experience from Danish wastewater treatment plants. *Water Science and Technology,* **42;** 65-72.

Oles, J., N. Dichtl and H. Niehoff (1997) Full scale experience of two stage thermophilic/mesophilic sludge digestion. *Water Science and Technology*, **36:** 449-456.

Page, A.L., T.J. Logan, and J.A. Ryan (1999) *Land Application of Sludge*, Lewis Publishers: Chelsea, Michigan.

Parkin, G.F. and W.F. Owen, (1986) Fundamentals of anaerobic digestion of wastewater sludges. *Journal of Enviornmetnal Engineering,* **112;** 867-920.

Rimkus, R.R., J.M. Ryan and E.J. Cook (1982) Full-scale thermophilic digestion at the West-Southwest Sewage Treatment Works, Chicago, Illinois. *Journal of Water Pollution Control Federation*, **54:** 1447-1457.

Rittmann, B.E., and P.L. McCarty (2001) *Environmental Biotechnology: Principles and Applications*, McGraw-Hill Higher: Boston.

Schafer, P.L. J.B. Farrell, G. Newman and S. Vandenburgh (2003) Advanced anaerobic digestion processes, *Water Environment and Technology*, **15:** 38-44.

Schwartzbrod, L. (1995) *Effect of Human Viruses on Public Health Associated with the Use of Wastewater and Sewage Sludge in Agriculture and Aquaculture*, EOS95-19, World Health Organization: Geneva.

Stumm, W. and J.J. Morgan (1996) *Aquatic Chemistry*. John Wiley and Sons: New York.

U.S. Environmental Protection Agency. (1979) Title 40 CFR Part 257 *Criteria for Classification of Solid Waste Disposal Facilities and Practices*, Washington DC.

U.S. Environmental Protection Agency (1994) A Plain English Guide to the EPA Part 503 *Biosolids Rule*. Office of Wastewater Management. EPA/832/R-93/003.

U.S. Environmental Protection Agency (1995) 40 CFR Parts 403 and 503, **60:** 54764.

U.S. Environmental Protection Agency (1999) *Control of Pathogens and Vector Attraction in Sewage Sludge*, EPA/625/R-92/013 Revised 1999, U.S. Environmental Protection Agency, Washington, DC.

World Health Organization (2000) Almost Half the World's People Have No Acceptable Means of Sanitation. PR-2000-73.

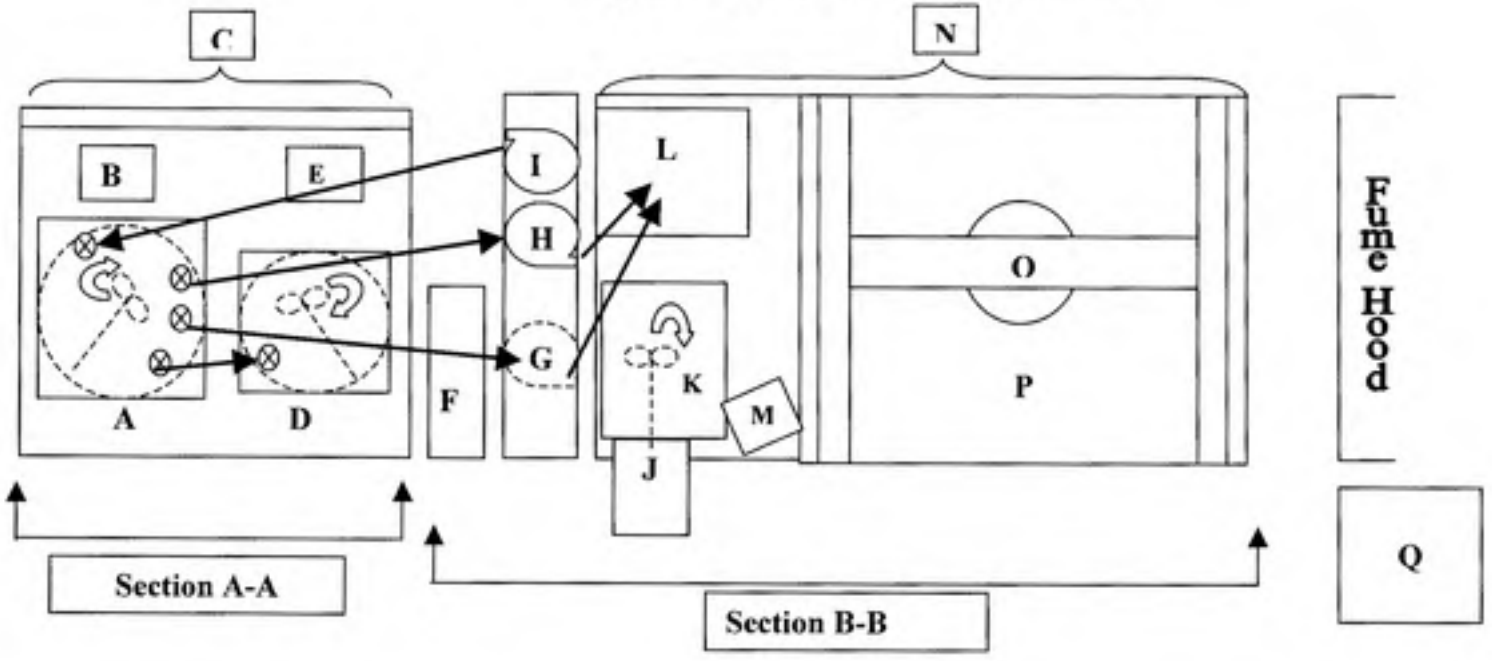
Young, H.D. (1962) *Statistical Treatment of Experimental Data*. McGraw-Hill: New York.

Zabranska, J. M. Dohanyos, P. Jenicek and J. Kutil (2002) The contribution of thermophilic anaerobic digestion to the stable operation of wastewater sludge treatment, *Water Science and Technology,* **46:** 447-453.

Zabranska, J., J. Stepova, R. Wachtl, P. Jenicek and M. Dohanyos (2000) The activity of anaerobic biomass in thermophilic and mesophilic digester at different loading rates. *Water Science and Technology,* **42:** 49-56.

6. APPENDICES

Appendix 1. Instrumentation Schematics



Legend:

A- 20-L CMFR
B- Constant Temperature Circulator 1
C- 125-L Water Bath
D- 4.5-L CMBR
E- Constant Temperature Ciruculator 2
F- Chilled Constant Temperature Ciruculator
G- Peristaltic Overflow Pump (P5)
H- Peristaltic Effluent Pump (P4)
I- Peristaltic Feed Pump (P3)
J- Feed Tank Mechanical Mixer
K- 20-L Feed Tank
L- 20-L Effluent Tank
M- BOD-Cubator
N- Storage cooler
O- Reservoir Sump Pump Mixer (P6)
P- 145L Sludge Reservoir
Q- Gas Chromatograph

Figure A1.1. Plan View, Tubing for sludge and effluent biosolids is indicated with bold arrows

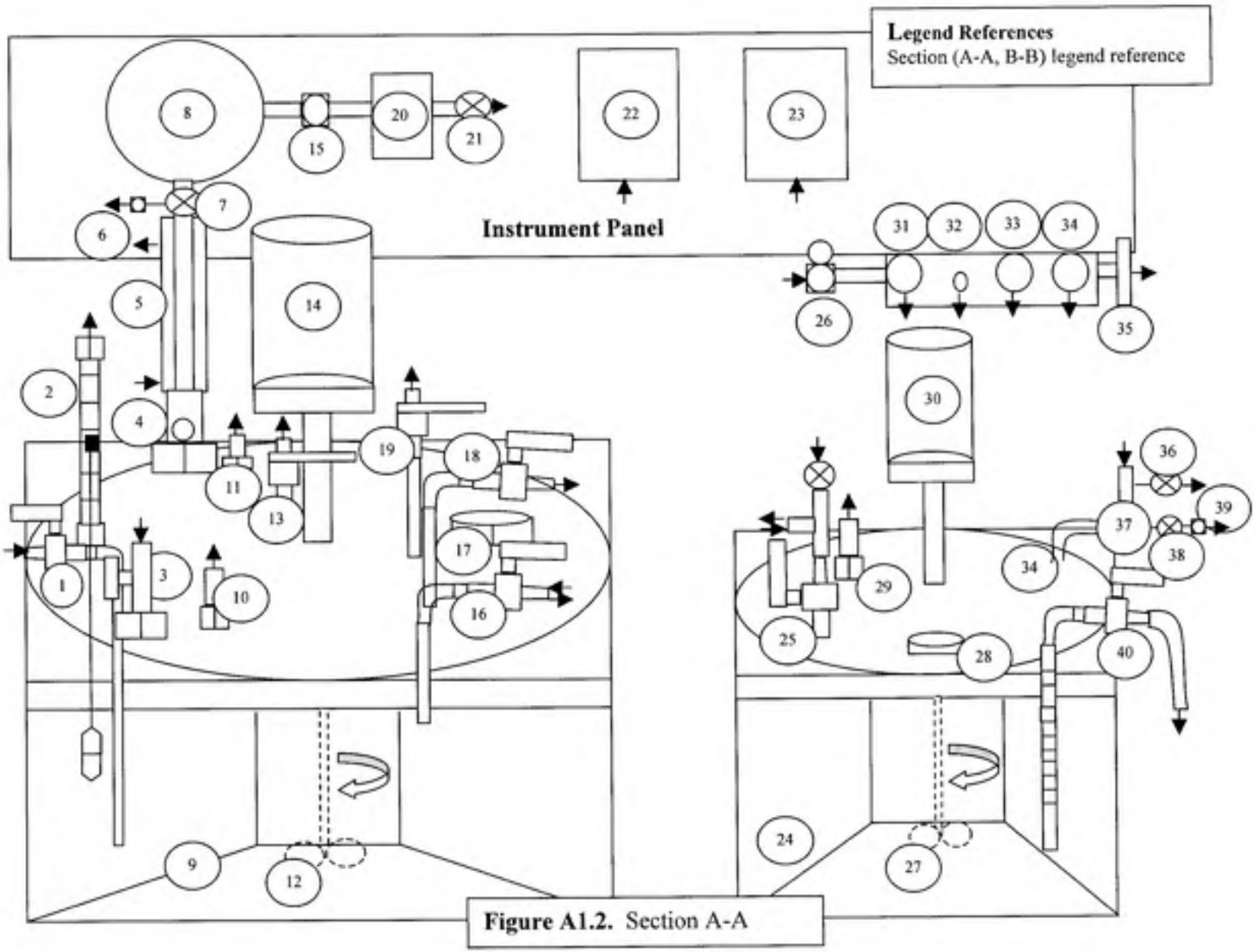


Figure A1.2. Section A-A

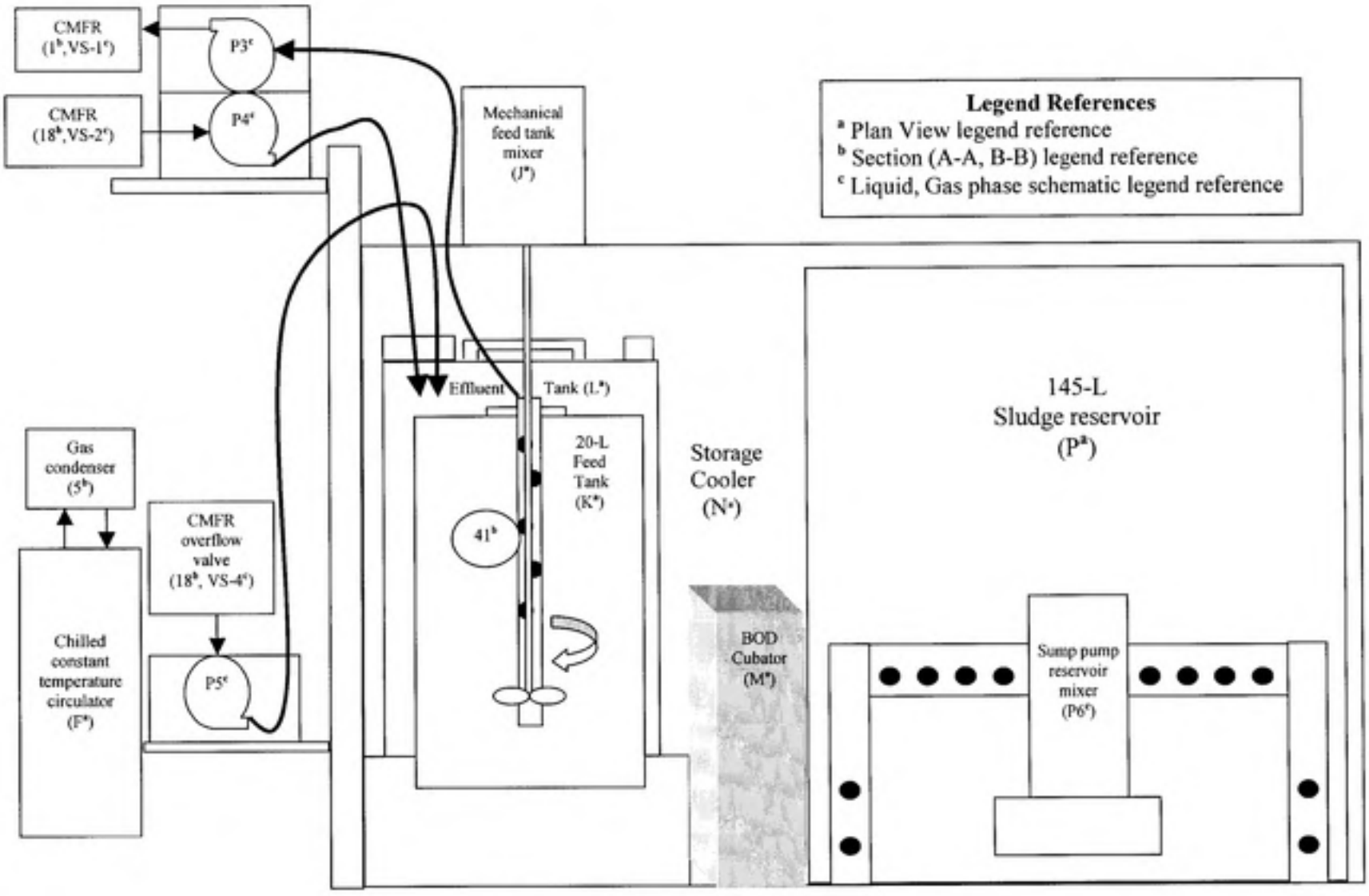


Figure A1.3. Section B-B

Legend A1.1. Sections (A-A, B-B)

1- Feed port for CMFR (VS-1), ¼" brass needle valve, allows entry of feed sludge from feed tank to CMFR, tubing outlet set near the bottom of the reactor to reduce short circuiting

2- Volume gauge for CMFR, calibrated and scored acrylic tubing, thin metal rod with a plastic float one end and a magnetic switch at the top to trigger the overflow pump at a volume of 20.4-L, measures CMFR biosolids volume

3- Argon purge valve for CMFR (VA-8), ¼" brass needle valve, connects gas manifold to CMFR

4- Effluent gas port for CMFR with one way, bubble breaker ball valve, protects gas phase instrumentation from biosolids

5- Condenser, jacketed stainless steel, circulates cold water from the chilled constant temperature circulator around CMFR effluent gas tygon tubing

6- Sampling post for gas chromatograph, connects CMFR to GC (Q)

7- Effluent gas shut off valve for CMFR (VA-9), ¼" nylon ball valve, severs gas flow from pressure gauge and gas flow meter

8- Pressure gauge (P_{G2}) for CMFR, Scientific Technologies Inc., model PG1000, measured pressure in CMFR

9- Cylindrical, three-vane baffle for CMFR, stainless steel, deters vortex mixing

10- Electric thermometer YSI Type-401, directly contacting CMFR biosolids, signaled to electric thermistor 1 (22)

11- Over pressure switch for CMFR, sensed high pressure with diaphragm switch and signaled to feed pump (P3) auxillary input

12- Impeller for CMFR mixer, three blade impeller

13- Effluent gas relief valve for CMFR (VA-10), ¼" nylon ball valve, vented CMFR gas

14- Mixer motor for CMFR, Leeson 44 W (1/17 hp), 300 rpm max., 90 VDC motor, drives impeller at variable speeds

15- Back pressure regulator for CMFR (R3), Airtrol RV5300-3.5, maintains positive pressure in CMFR

16- Biosolids transfer port for CMFR (VS-3), 3/8" stainless steel ball valve, connects CMFR to CMBR for inactivation tests

Legend A1.1. Sections (A-A, B-B)

17- Auxiliary port for CMFR, 1 ½" nut tightens, scored plastic meBRane, and rubber gasket, provides safety control for over pressurization and access to biosolids

18- Overflow valve for CMFR (VS-4), 3/8" stainless steel ball valve, stainless steel tubing inlet set at a height corresponding to 19.3-L, connects CMFR to effluent tank (L)

19- Effluent valve for CMFR (VS-2), 3/8" stainless steel ball valve, set in the upper section to deter short circuiting, connects CMFR to effluent tank (L)

20- Gas flow meter for CMFR, Omega FMA-1600, measures CMFR effluent gas flow rate

21- Gas flow meter isolation valve (VA-17), ¼" nylon ball valve, connects gas flow meter (20) and fume hood

22- Electric thermistor 1 for CMFR, Cole-Palmer Digi-Sense ThermoLogR, signals from electric thermometer (10) to data acquisition system

23- Electric thermistor 2 for CMBR, Cole-Palmer Digi-Sense ThermoLogR, signals from electric thermometer (29) to data acquisition system

24- Cylindrical three-vane baffle, stainless steel

25- Biosolids transfer valve from CMFR to CMBR (VS-5), with argon purge valve (VA-7) from gas manifold (VA-14) to CMBR (VS-5)

26- Argon regulator for gas manifold (R2) with pressure gauge (P_{g1}), maintains and measures positive pressure from Argon source tank to gas manifold

27- Mixer impeller for CMBR, three blade impeller

28- Pathogen spiking port for CMBR, threaded, Teflon plug

29- Electric thermometer for CMBR, YSI Type 401, directly contacting CMBR biosolids, signaled to electric thermistor 2 (23)

30- Mixer motor for CMBR, Dayton 93 W (1/8 hp), 1800 rpm max., 90 VDC motor

31- Argon purge for CMFR, directly connects gas manifold to argon purge valve on CMFR (VA-8), 3/8" stainless steel ball valve

32- Sludge transfer line, argon pressurization metering valve from gas manifold (VA-14), ¼" brass needle valve, to transfer line (25) between CMFR and CMBR

33- Argon purge valve from gas manifold (VA-15), ¼" brass needle valve, to sludge reservoir (P)

Legend A1.1. Sections (A-A, B-B)

34- Argon purge valve from gas manifold (VA-11), ¼" brass needle valve, to CMBR (VA-12), ¼" nylon ball valve

35- Argon purge valve from gas manifold (VA-16), ¼" brass needle valve, to unconnected tubing for general purging use

36- Effluent gas relief valve for CMBR (VA-12), ¼" nylon ball valve, connects CMBR to fume hood

37- Pressure gauge for CMBR (P_{G3}), measures pressure in CMBR

38- Effluent gas regulated relief valve for CMBR (VA-13), ¼" nylon ball valve, connects CMBR to back pressure regulator (39)

39- Back pressure regulator for CMBR (R4), maintains positive pressure in CMBR

40- Sampling port for CMBR (VS-6), 3/8" stainless steel ball valve with calibrated, scored stainless steel tubing, allows for drawing biosolids samples during inactivation tests

41- Feed tubing conduit, PVC tubing with drilled holes, holds feed tubing in feed sludge

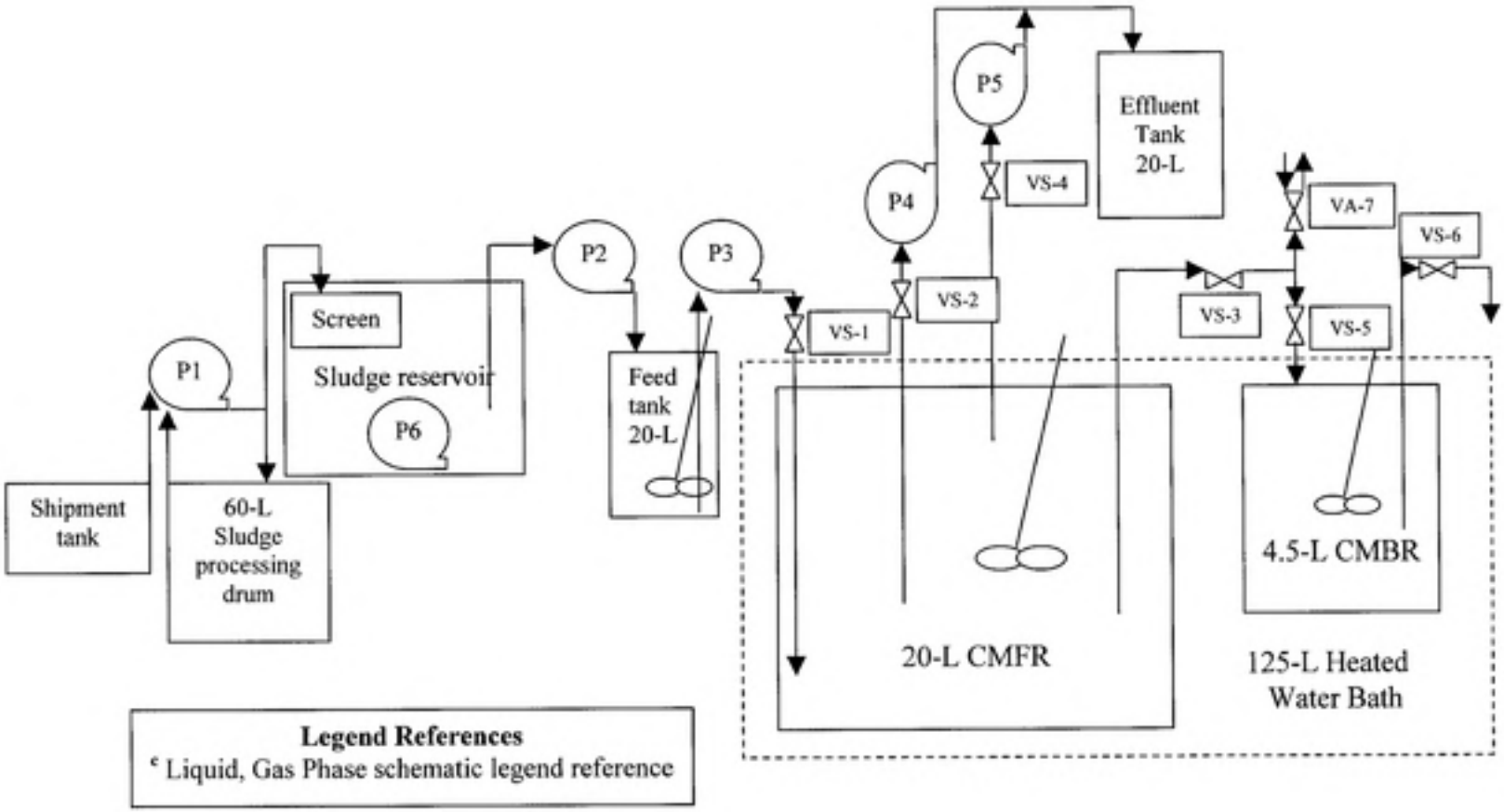


Figure A1.4. Liquid Phase Schematic

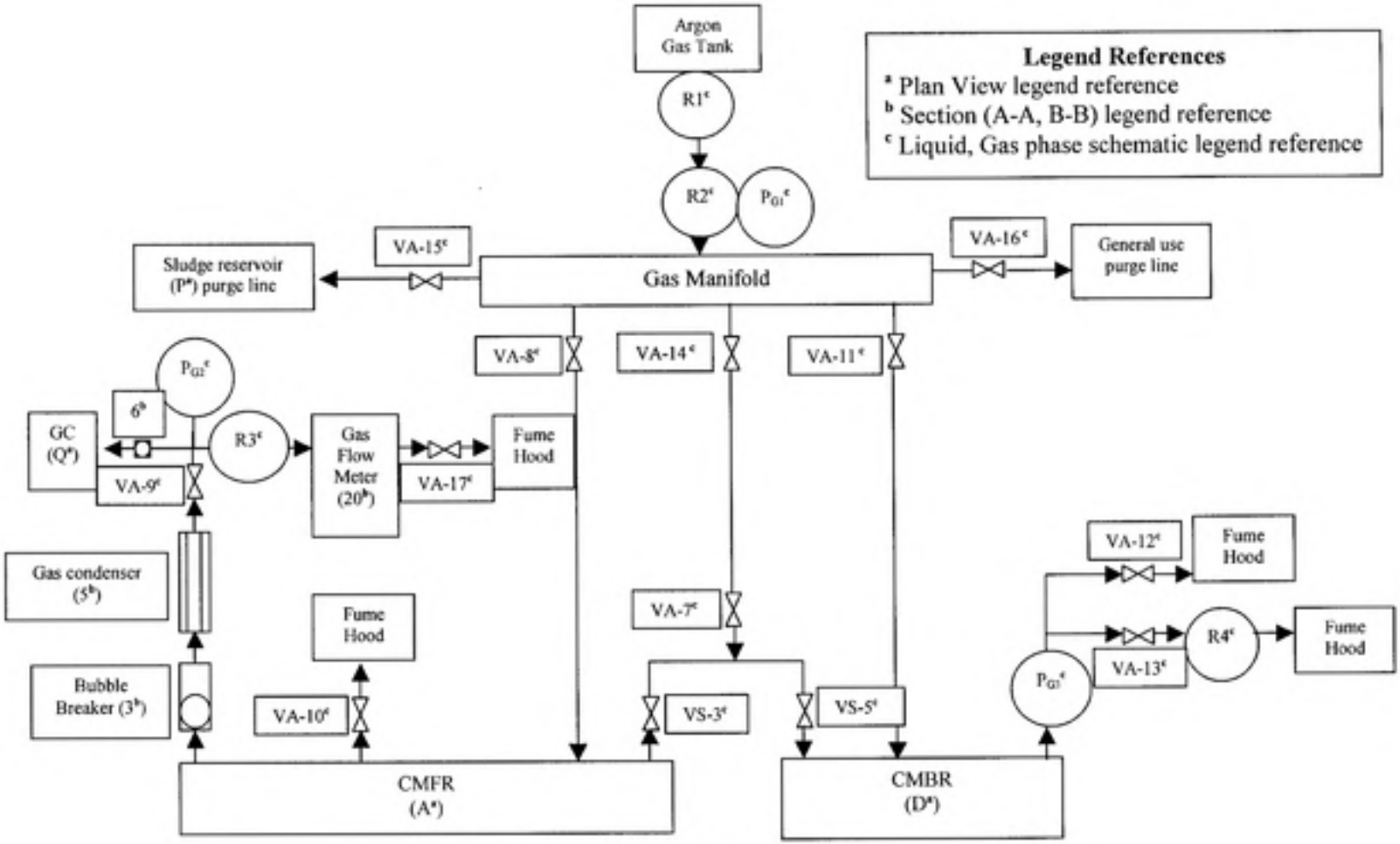


Figure A1.5. Gas Phase Schematic

Legend A1.2. Liquid and Gas Phase Schematics

Pumps (P)

P1: Macerator pump – Jabsco 12 gpm, 12 VDC, macerator pump, model 18590-1000 – used for processing raw sludge and transferring to sludge reservoir

P2: Sludge transfer pump – Masterflex, 0-600 rpm, variable speed peristaltic pump – used for transferring sludge from the sludge reservoir to the feed tank using LS-35 Norprene tubing

P3: Feed pump – Masterflex, 0-60 rpm, computer drive peristaltic pump – used for feeding sludge from the feed tank to the continuous reactor using LS-35 Norprene tubing

P4: Effluent pump – Masterflex, 0-60 rpm, computer drive peristaltic pump – used for drawing biosolids from the continuous reactor to the effluent tank using LS-35 Norprene tubing

P5: Overflow pump – Masterflex 0-100 rpm variable speed reversible peristaltic pump – used for drawing excess bisolids from the continuous reactor to the effluent tank using LS-35 Norprene tubing

P6: Sludge reservoir mixing pump – Grundfos 0.3 kW [0.4 hp] Type 316 stainless steel/polypropylene sewage ejector pump- used for mixing the sludge reservoir contents for 30 sec. per hour

Valves – Liquid phase (VS), Gas phase (VA)

VS-1: Feed valve – 3/8" stainless steel ball valve

VS-2: Effluent valve - 3/8" stainless steel ball valve

VS-3: Biosolids transfer valve on continuous reactor - 3/8" stainless steel ball valve

VS-4: Overflow valve on continuous reactor - 3/8" stainless steel ball valve

VS-5: Biosolids transfer valve on batch reactor - 3/8" stainless steel ball valve

VS-6: Biosolids sampling valve on batch reactor – 3/8" stainless steel ball valve

VA-7: Biosolids transfer line purge valve on manifold side of T-joint for batch and continuous reactor – ¼" nylon ball valve

VA-8: CMFR gas purge valve – ¼" brass needle valve

VA-9: CMFR effluent gas shut-off valve – ¼" nylon ball valve

VA-10: CMFR gas relief valve - ¼" nylon ball valve

VA-11: CMBR gas purge valve – ¼" brass needle valve

VA-12: CMBR purge gas relief valve – ¼" nylon ball valve

Legend A1.2. Liquid and Gas Phase Schematics

VA-13: CMBR effluent gas relief valve - ¼" nylon ball valve

VA-14: Sludge transfer line metering valve – ¼" brass needle valve

VA-15: Sludge reservoir purge valve - ¼" brass needle valve

VA-16: General purpose argon purge valve - ¼" brass needle valve

VA-17: Digester flow meter isolation valve - ¼" nylon ball valve

Gas regulators (R)

R1: Argon regulator for source tank

R2: Argon regulator for gas manifold

R3: CMFR gas back pressure regulator

R4: CMBR gas back pressure regulator

Pressure gauges (P_G)

P_{G1}: Pressure gauge for gas manifold

P_{G2}: Pressure gauge for CMFR - Scientific Technologies Inc., model PG100

P_{G3}: Pressure gauge for batch reactor

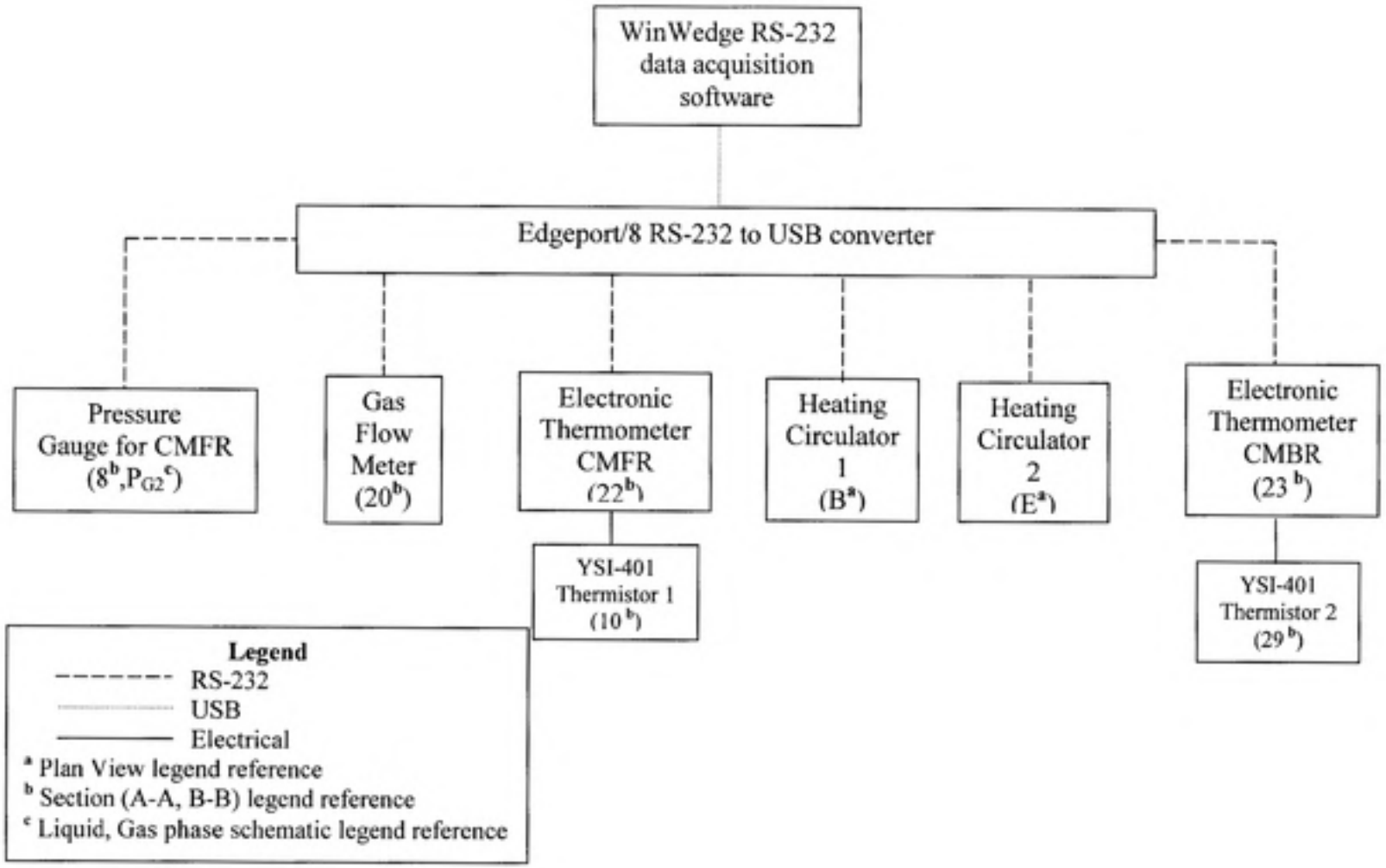
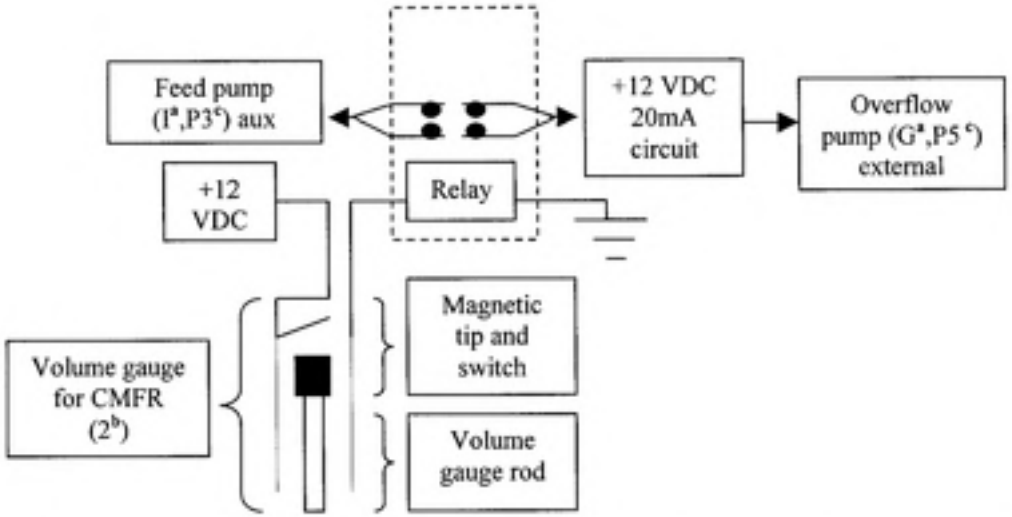
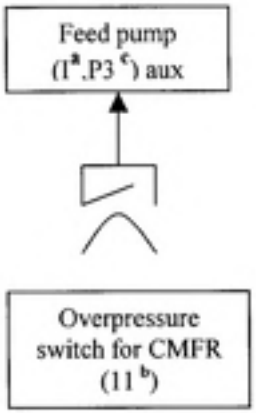


Figure A1.6. Data Acquisition and Control Schematic-A



B.1. Magnetic switch and circuitry for volume gauge



B.2. Diaphragm overpressure switch

Legend

[a] Plan View legend reference
[b] Section (A-A, B-B) legend reference
[c] Liquid, Gas phase schematic legend reference

Figure A1.7. Data Acquisition and Control Schematic-B

Appendix 2. Experiment Chronology

Table A2.1. Operating Condition History

Operating Condition			
Sludge Source	**Temperature (°C)**	**Date**	**Event**
SCWRF	55	7/26/02	digester seeded with biosolids from OWASA first-stage thermophilic anaerobic digester
		8/30/02	reached desired residence time
		9/7/02	first batch test
		9/14/02	second batch test
		11/15/02	digester re-seeded [a]
		12/6/02	reached desired residence time
		12/14/03	first inactivation rate test
		1/4/03	second inactivation rate test
SCWRF	53	1/5/03	changed operating temperature to 53 °C
		1/18/03	first batch test
		1/25/03	second batch test
		2/1/03	first inactivation rate test
		2/8/03	second inactivation rate test
SCWRF	51	2/10/03	operating temperature changed to 51 °C
		2/21/03	digester re-seeded [b]
		2/25/03	reached desired residence time
		3/1/03	first inactivation rate test
		3/8/03	second inactivation rate test
		3/15/03	first batch test
		3/22/03	second batch test
OWASA	53	3/25/03	feed source changed to OWASA fermented primary sludge; operating temperature changed to 53 °C
		4/5/03	first batch test
		4/12/03	second batch test
WLSSD	53	4/14/03	feed source changed to WLSSD sludge
		4/26/03	first batch test
		5/3/03	second batch test
SCWRF	49	5/7/03	operating temperature changed to 49 °C
		5/9/03	feed sludge changed to SCWRF
		5/10/03	inactivation rate test

[a] Digester was re-seeded after the cause of failures (inhibitory substances in the stock materials used to supplement the feed sludge with *Ascaris* and poliovirus) was identified and corrected.

[b] Digester failure occurred after a stock of poliovirus that had not been washed to eliminate antibiotics was used to supplement the feed sludge on 2/18/03.

Table A2.2. Date Ranges for Sampling at Each Operating Condition [a]

Operating Condition		Date Range for Sampling Continuous Digester	
Sludge Source	**Temperature (°C)**	**Feed**	**Effluent**
SCWRF	55	8/30/02 – 9/13/02 12/6/02 – 12/11/02	9/3/02 – 9/13/02 12/9/02 – 12/13/02
SCWRF	53	1/7/03 – 2/7/03	1/13/03 – 2/7/03
SCWRF	51	2/28/03 – 3/22/03	3/4/03 – 3/22/03
OWASA	53	3/28/03 – 4/14/03	3/31/03 – 4/14/03
WLSSD	53	4/16/03 – 4/30/03	4/20/03 – 5/2/03

[a] Data from samples collected on dates outside this range were not used in data summaries.

Appendix 3. Flow Rate Data

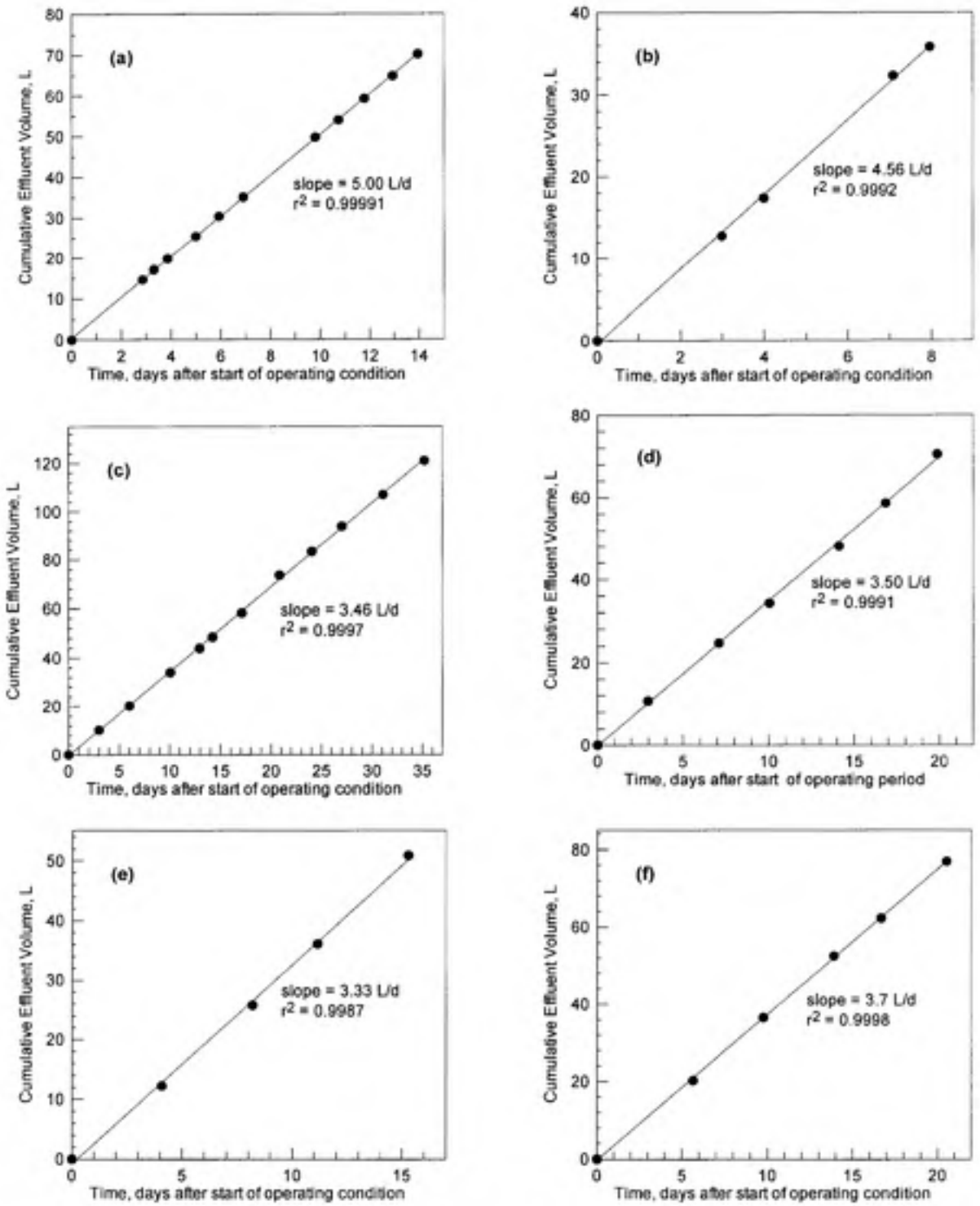


Figure A3.1. Cumulative effluent volume vs. time for operation at **(a)** 55 °C, SCWRF sludge, period 1; **(b)** 55 °C, SCWRF sludge, period 2; **(c)** 53 °C, SCWRF sludge; **(d)** 51 °C, SCWRF sludge; **(e)** 53 °C, OWASA sludge; and **(f)** 53 °C, WLSSD sludge

Appendix 4. Temperature Calibration Records

Table A4.1. Thermistor Calibration Record [a]

Date	Temperature (°C) [b]	Thermistor 1 Reading (°C)		Thermistor 2 Reading (°C)	
		Before Calibration	After Calibration [c]	Before Calibration	After Calibration [c]
8/16/02	55.10	54.95	55.05	4.98	55.08
9/6/02	55.20	55.02	55.22	54.96	55.16
9/13/02	55.10	55.03		55.09	
9/25/02	53.00	53.06		53.00	
10/18/02	53.10	53.06		53.10	
10/20/02	53.10	55.18	52.54		
10/21/02	53.10	50.28 [d]	53.08	49.59 [d]	53.09
11/2/02	53.10	52.91	53.06		
11/17/02	55.10	55.35	55.15	55.23	55.13
11/26/02	55.10	55.00	55.09		
12/9/02	55.10	56.46	55.06		
12/10/03	55.13	53.57 [d]	55.07	54.97 [d]	55.07
12/20/03	55.10	55.14	55.12	55.15	55.14
12/30/02	55.00	54.97		54.98	
1/17/03	53.10			53.07	
1/31/03	53.10			53.05	
2/3/03	53.00			52.88	53.00
2/7/03	53.10	52.98	53.08	53.09	
2/25/03	51.20	51.15		51.50	
3/7/03	51.00	50.96		50.96	
3/14/03	51.00	50.98		50.96	
3/20/03	51.00	51.01		50.98	
4/4/03	53.00	53.00		52.95	
4/11/03	53.00	53.02		52.92	53.01
4/24/03	53.05	53.03		53.00	
5/2/03	53.10	53.02	53.12	52.94	53.15

[a] Thermistor 1 was used in the continuous reactor and Thermistor 2 in the batch reactor.

[b] Water bath temperature measured with the NIST-traceable thermometer.

[c] Reading is shown only if adjustment was made.

[d] Calibration of replacement thermistor.

Table A4.2. Constant-Temperature Circulator Calibration Record

Date	Temperature (°C) [a]	Circulator 1 Reading (°C)		Circulator 2 Reading (°C)	
		Before Calibration	After Calibration [b]	Before Calibration	After Calibration [b]
8/16/02	55.10	55.1		55.1	
8/21/02	55.10	55.1		55.1	
8/29/02	55.30	55.1	55.3	55.2	55.3
9/6/02	55.20	55.2		55.2	
9/13/02	55.10	55.1		55.1	
9/25/02	53.00	53.0		53.0	
10/18/02	53.10	53.1		53.1	
10/21/02	53.10	53.1		53.1	
11/17/02	55.15	55.1		55.2	55.16
11/26/02	55.10	55.1		55.1	
12/10/02	55.13	55.1		55.0	55.1
12/20/02	55.10	55.1		55.2	55.1
12/30/02	55.00	55.1		55.0	55.1
1/9/03	53.10	53.1		53.1	
2/28/03	51.10	51.0		51.0	51.1
2/28/03	51.10	51.1		51.1	
3/20/03	51.00	51.1		51.1	
4/11/03	53.00	53.1		53.1	53.0
4/24/03	53.05	53.1		53.1	
5/2/03	53.10	53.1		53.1	

[a] Water bath temperature measured with the NIST-traceable thermometer.

[b] Reading is shown only if adjustment was made.

Appendix 5. Calibration of the Gas Flow Meter

The digital readout of the gas flow meter depends on the kinematic viscosity of the gas flowing through it. The meter is manufactured with settings for various pure gases, none of which matched the composition of digester gas (approximately one-third carbon dioxide and two-thirds methane). From experience, the setting for N_2O gave readouts closest to the actual digester gas flow rate. The meter was then calibrated at this setting by making repeated measurements of digester gas flow over a short interval with a bubble flow meter and simultaneously recording the average meter reading over that interval using the data acquisition software. Two calibration checks were conducted on 9/19/02 and a third check was conducted on 11/2/02. The correlation between meter reading and actual gas flow determined on 9/19/02 was used to quantify gas flow rates for the first operating period at 55 °C with sludge from the SCWRF. The correlation determined on 11/2/02 was used to quantify gas flow rates for all the remaining operating periods. The ratios of actual flow to meter reading for the tests on 9/19/02 were 1.13 ± 0.01 in both cases. The ratio of actual flow to meter reading for the test on 11/2/02 was 1.16 ± 0.02.

Results of the calibration measurements are shown in the tables below.

Table A5.1. Gas Flow Calibration Measurements for Test 1 on 9/19/02

Bubble Meter			
Bubble Travel Time (seconds) [a]	**Flow Rate (mL/min)**	**Average Flow Meter Reading**	**Ratio of Actual Flow to Meter Reading**
19.20	31.25	27.6	1.132
19.63	30.57	27.8	1.099
19.69	30.47	27.2	1.120
19.22	31.22	27.3	1.143
19.33	31.04	27.7	1.121
28.51 [b]	31.57	27.7	1.140
19.04	31.51	28.1	1.121
18.76	31.98	28.4	1.126
18.40	32.61	28.8	1.132
18.52	32.40	28.7	1.129
18.77	31.97	28.3	1.130

[a] Time for bubble to travel 10 mL except as indicated.

[b] Time for bubble to travel 15 mL.

Table A5.2. Gas Flow Calibration Measurements for Test 2 on 9/19/02

Bubble Meter			
Bubble Travel Time (seconds) [a]	**Flow Rate (mL/min)**	**Average Flow Meter Reading**	**Ratio of Actual Flow to Meter Reading**
19.98	30.03	26.9	1.116
20.04	29.94	26.9	1.113
19.60	30.61	26.8	1.142
19.86	30.21	27.0	1.119
19.52	30.74	27.1	1.134
20.38	29.44	26.1	1.128
20.19	29.72	26.2	1.134

[a] Time for bubble to travel 10 mL.

Table A5.3. Gas Flow Calibration Measurements for Test 3 on 11/2/02

Bubble Meter			
Bubble Travel Time (seconds) [a]	**Flow Rate (mL/min)**	**Average Flow Meter Reading**	**Ratio of Actual Flow to Meter Reading**
44.72	26.83	23.9	1.125
44.19	27.16	23.7	1.146
42.07	28.52	24.2	1.179
40.50	29.63	25.4	1.169
40.58	29.57	25.2	1.176
42.23	28.42	24.5	1.162
45.46	26.40	22.4	1.181
42.03	28.55	24.4	1.170
42.20	28.44	24.6	1.156
42.22	28.42	24.3	1.170

[a] Time for bubble to travel 20 mL.

Appendix 6. Summary Statistics on Replicate Analyses for Total and Volatile Solids

The coefficient of variation (or relative standard deviation) is the standard deviation of a data set divided by the mean of the data set. Coefficients of variation were calculated for each feed sludge and effluent biosolids sample analyzed in triplicate throughout the reported operating periods. Average and maximum values are summarized in Table A6.1

Table A6.1. Summary Data on Coefficients of Variation for Solids Analyses

		Coefficient of Variation	
Sample type	**No. of Samples**	**Average**	**Maximum**
TS in feed sludge	31	0.015	0.078
VS in feed sludge	31	0.016	0.075
TS in effluent	30	0.008	0.050
VS in effluent	29	0.009	0.019

Appendix 7. Analytical Balance Calibration Record

Table A7.1. Analytical Balance Calibration Record

Date	Calibration Weight (g)	Initial Reading (g)	Error (g)	Error Relative to 55 g Weight (g) [a]	Reading After Calibration (g)
9/9/02	100	100.0110	+ 0.0110	+ 0.0061	100.0008
4/23/03	200	199.9860	- 0.0140	- 0.0039	200.0001
4/30/03	200	199.9980	- 0.0020	- 0.0001	199.9997
5/2/03	200	200.0012	+ 0.0012	+ 0.0033	200.0002
5/12/03	200	199.9954	- 0.0046	- 0.0013	200.0000

[a] Approximate weight of a crucible used in the analysis of total and volatile solids.

Appendix 8. Calibration Checks of Acid Titrant Used for Alkalinity Measurements

Table A8.1. Calibration checks on acid titrant

Date	Measured Concentration (eq/L)
6/24/02	0.496
7/19/02	0.496
9/30/02	0.499
10/1/02	0.489 [a]
1/23/03	0.489
6/30/03	0.492

[a] New batch of acid.

Appendix 9. Correlation of VFAs Measured by Titration vs. GC Analysis

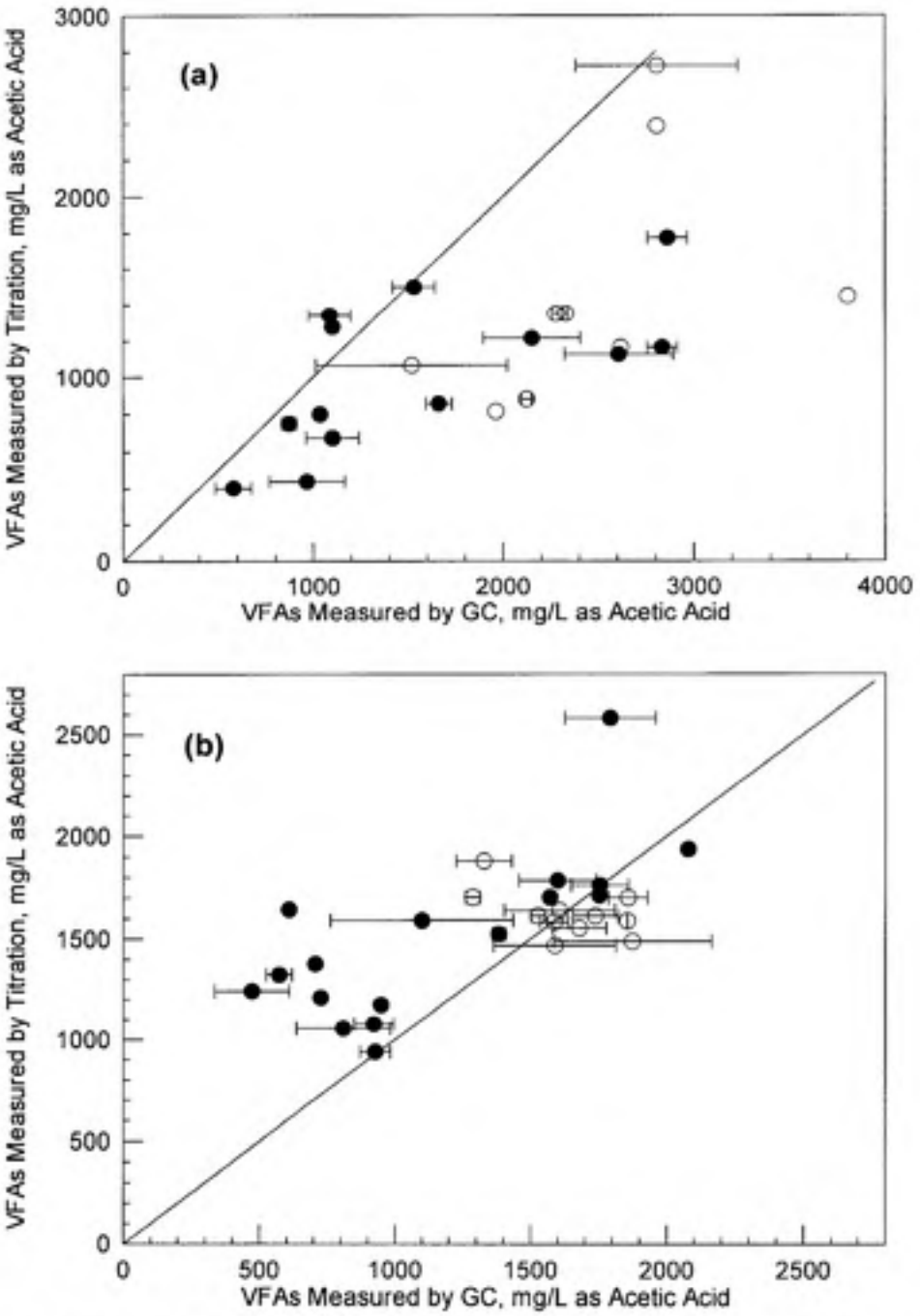


Figure A9.1 Total VFAs (as acetic acid) measured by titration vs. total VFAs measured by gas chromatography for **(a)** feed sludges and **(b)** effluent biosolids. The open circles represent samples that were analyzed by GC for storage periods greater than two months (all samples between 8/30/02 and 12/11/02). Error bars are standard deviations from GC analysis of replicate samples (usually duplicates). The diagonal lines represent a 1:1 correlation between the two methods.